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The ontogeny of the postcranial skeleton in saddle-back tamarins, *Leontocebus fuscicollis* and callimicos, *Callimico goeldii* (Callitrichidae, Primates)

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Abstract: Ontogenetic studies of callitrichid anatomy are limited to research focused mainly on postcranial skeleton of adults. The goal of this study is to compare the ontogeny of postcranial skeletal development in Goeldi's monkeys (i.e., callimico; *Callimico goeldii*) with the corresponding data on saddle-back tamarins (*Leontocebus fuscicollis*). The intermembral, humerofemoral, brachial, crural, and ulna-radius indices of callimicos and saddle-back tamarins were calculated and compared among different age classes in order to assess the implications for their ecology and behavior. Ontogenetic trajectories, including age at growth cessation, were also calculated. It is shown that for a given hindlimb length, *L. fuscicollis* has longer forelimbs compared to *C. goeldii*, maintaining this proportion across all age classes. A relatively elongated forelimb observed in *L. fuscicollis* may have a mechanical role in reducing the force of impact when landing on large vertical substrates. In contrast, hindlimb length and pattern of hindlimb development (such as derived features of the ankle that enhance stability) in callimicos appear to play a critical role in propulsion during trunk-to-trunk leaping. These differences may affect niche partitioning, foraging strategies, and substrate use.

Key words: allometry, growth, limb proportions, New World primates, ontogeny, postcranial skeleton.

Original article

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Introduction

Research studies on ontogeny (i.e., the course of growth and development of individuals to maturity) and allometry (i.e., the study of size and its implications) in individual primate species facilitate better understanding of the evolutionary adaptive histories of the Primate order (Fleagle 1985; Shea 1995; Marroig and Cheverud 2009; for hominid primates: Gould 1977; Leigh 1996a; Nelson and Thompson 1999). Thus, research on the postcranial skeleton, and on the fore- and hindlimbs specifically, offers an opportunity to address questions concerning variation in locomotor patterns and body size among primates (Jungers 1985; Falsetti et al. 1993; Leigh 1996b; Leigh and Shea 1995). In this sense, the diversity of primate limb skeletons is related to natural selection and may reflect the variability in the use of different forest substrates (Chiu and Hamrick 2002).

Regarding the Callitrichidae family (Rylands et al. 2016), there have been few studies that emphasized the importance of postcranial development (e.g., Glassman 1983; Falsetti and Cole 1992). For example, Bicca-Marques et al. (1997, 1998) and Bicca-Marques (1999) compared data on hand morphology among different species of callitrichids, illustrating the relevance of hand shape in relation to their feeding ecology and niche partitioning. However, as indicated by Falsetti and Cole (1992), these data were mainly derived from adult individuals. In contrast, only few studies have taken into account the relevance of studying the ontogeny among callitrichids in order to understand their implications regarding behavioral ecology and positional behavior of this group (Falsetti and Cole 1992; Garber and Leigh 1997; Garber and Leigh 2001a).

Positional and foraging behavior of callitrichids is adapted to the use and preference of low forest substrates (Terborgh 1983; Yoneda 1984; Garber and Teaford 1986; Heymann 1997; Garber and Leigh 2001a, 2001b). The aim of this study is to describe the ontogeny of the postcranial skeleton of callimicos (*Callimico goeldii*) and saddle-back tamarins (*Leontocebus fuscicollis*), and to determine its implications for understanding the development of their positional behavior. These two callithichid taxa are reported to be the most frequent trunk-to-trunk leapers within this primate group (Garber and Leigh 1997; Garber and Leigh 2001) and are sympatric in the wild. This paper aims to provide further information on the postcranial skeleton proportions of the Callithrichidae following the comprehensive work of Davis (2002).

This research has the following objectives: (i) to calculate and compare proportional indices of postcranial skeletons in *C. goeldii* and *L. fuscicollis* immatures/matures; (ii) to determine whether there are intra- and interspecific differences or similarities between both species and between age-classes in terms of their ontogenetic and allometric histories; (iii) to reconstruct the ontogeny of these primates using fore- and hindlimbs; and (iv) to evaluate the relationships between the morphology of the postcranial skeleton and the behavior of both species. Relating morphology and behavior offers insight into the ecological adaptability of these New World primates.

Material and Methods

The saddle-back tamarins (*Leontocebus fuscicollis*) specimens used in this study consisted of 22 females and 18 males (total 40 individuals) including 18 immature (<190 days) and 22 mature (>190 days) individuals (according to the age classes provided by Garber and Leigh (1997)). Data on the exact age of death have been recorded in all examined individuals. The skeletal research collection is housed in the Laboratory of Primate Biology of the Department of Anthropology at the University of Illinois at Urbana-Champaign, USA. These saddle-back tamarin (*L. fuscicollis*) skeletons were curated and obtained from the Department of Anthropology at the University of Tennessee-Knoxville, USA and came from the Marmoset Research Center, Oak Ridge Associated Universities, USA. The original tamarin colony was created in the 1960s by N. Gengozian (1969) and used for medical studies. The skeletons used in this study were sampled from captive-born *L. fuscicollis* individuals belonging to three different subspecies (*illigeri*, *nigrifrons*, and *lagonotus*) and their hybrids. As suggested by Garber and Leigh (1997), the small number of *L. fuscicollis* subspecies used in this study could be a limitation. Although the sample size is ample for this genus, it is not large enough to be compared at the subspecific level. The data are pooled at the species level.

The callimicos (*Callimico goeldii*) skeleton collection is maintained by the Barbara E. and Roger O. Brown Primate Research Facility in the Division of Mammals at the Field Museum of Natural History, Chicago, USA. The specimens, eight females and 14 males (totaling 22 individuals), included six immature and 16 mature individuals. All individuals were captive-born at the Brookfield Zoo (Chicago). The ages were recorded and provided by M. Wanerke (1998, 2003: pers. comm.). The *C. goeldii* colony was founded in 1977 to maintain a long-term successful breeding project for this rare New World primate (Beck et al. 1982; Sodaro 2000; see also Palacios et al. 2021).

In order to pursue the objectives of this work, the maximum lengths of the femur (FML), tibia (TML), humerus (HML), radius (RML), and ulna (UML) were measured using the diaphyseal lengths. The measurements were done using a digital sliding caliper, the Mitutoyo™ 500–197, graduated to 0.01 mm. Data from males and females were pooled; there is no significant sexual dimorphism in this primate group (Hershkovitz 1977; Cole et al. 1988; Hanihara and Natori 1988, estimated by standard deviation in this study). While examining questions of ontogeny, the data were analyzed separately between species and age classes. Immature individuals show signs of ossification, as reported for callitrichid infants by Hofmann et al. (2007). The intermembral, humerofemoral, brachial, crural, and ulna-radius indices were calculated for both primates and age classes, considering the average captive adult body weights (Jungers 1985: 350) (for *L. fuscicollis* body weight [= 414.5 g]: Leigh 1994: 25 and for *C. goeldii* body weight [= 607 g]: Wanerke 2003: pers. comm.). Measurements were collected on non-pregnant and healthy animals.

Statistical analysis was performed using the SYSTAT® software package and Microsoft-Excel®. Ontogenetic data were graphically represented for both callitrichid species. An analytical comparison was done using conventional least squares regression analysis. Furthermore, to contrast pairs of variables, *t*-tests were conducted to establish potential differences among the indices of both primates and age classes.

Results

Figure 1 shows absolute differences in the postcranial maximum length (ML) of both callimicos and saddle-back tamarins. The postcranial proportion indices were calculated in order to compare intra- and interspecific variation in shape. As shown in Table 1, no significant differences were observed in any of the measurement taken from immature individuals of *Callimico goeldii* and *Leontocebus fuscicollis*. However, when comparing the ML proportion indices among mature individuals, *L. fuscicollis* shows significantly (99%, p<0.0001) higher intermembral and humerofemoral indices compared to *C. goeldii*. In contrast, *C. goeldii* has significantly higher values of the ulna-radius index compared to *L. fuscicollis* (Table 1; Fig. 2 shows statistically significant limb proportion indices). The species-specific differences of mature individuals are also indicated in several forelimb and hindlimb indices; in all cases, callimicos values are higher than saddle-back tamarins (Table 1).

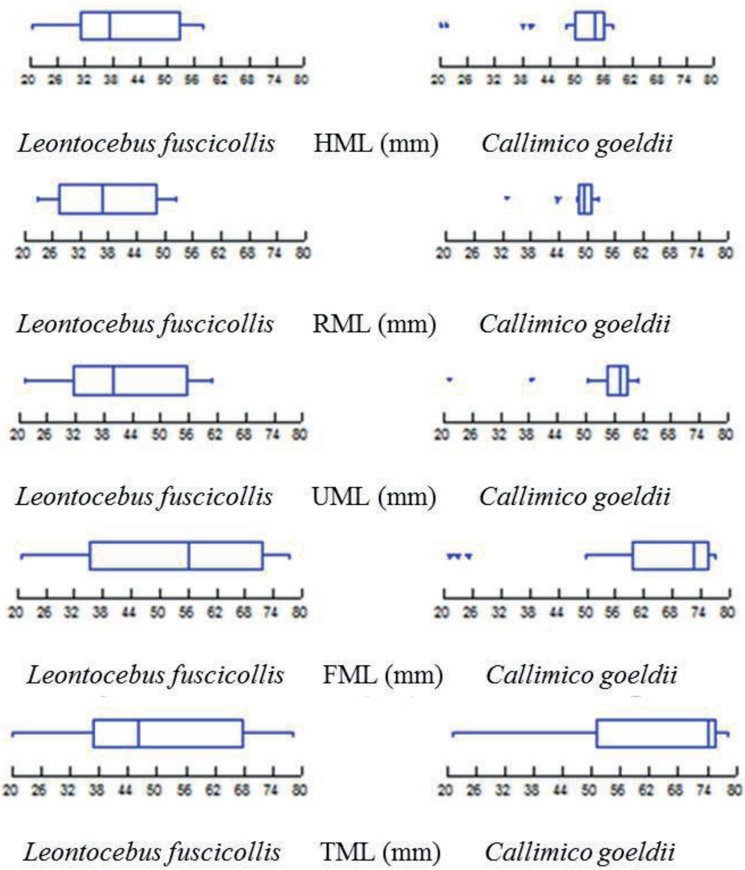

Fig. 1. Box plots for comparison of postcranial skeleton lengths (mm) between mature *Callimico goeldii* and *Leontocebus fuscicollis*

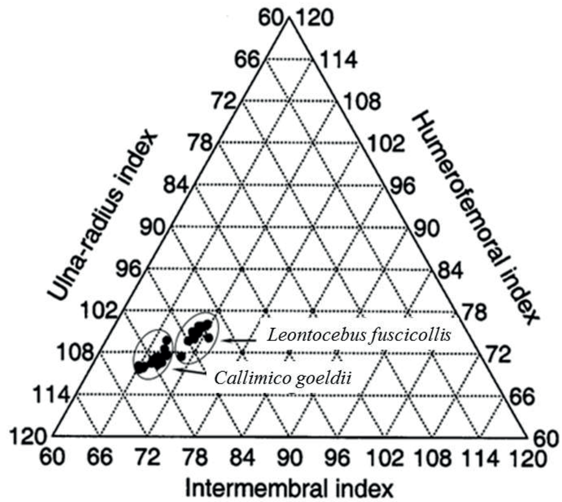

Fig. 2. Triangular plot of the significantly different relative limb proportion (indices) between mature *Callimico goeldii* and *Leontocebus fuscicollis*

Table 1. Comparison of postcranial skeleton proportions (indices) between immature and mature *Callimico goeldii* and *Leontocebus fuscicollis*

	Immature								
	Callimico goeldii			***Leontocebus fuscicollis***					
	n	**Mean**	**S. D.**	**n**	**Mean**	**S. D.**	***p*-value**	**d.f**	
Intermembral index	6	79.015	7.109	17	82.961	4.588	0,021	3	*L. f.>C. g.*
Humerofemoral index	4	80.701	8.839	13	90.122	4.091	0,054	3	L. f.>C. g.
Brachial index	6	90.191	11.037	18	89.268	4.096	0,780	5	*C. g.>L. f.*
Crural index	4	96.762	3.914	13	105.802	3.416	0,043	3	*L. f.>C. g.*
Ulna/radius index	6	111.206	6.167	18	113.733	3.784	0,791	5	L. f.>C. g.

	Mature								
	Callimico goeldii			***Leontocebus fuscicollis***					
	n	**Mean**	**S. D.**	**n**	**Mean**	**S. D.**	***p*-value**	**d.f**	
Intermembral index	16	70.073	1.485	21	77.424	1.441	0,000	12	*L. f.>C. g.**
Humerofemoral index	16	74.576	2.121	13	83.163	2.992	0,000	12	*L. f.>C. g.**
Brachial index	16	90.292	3.230	21	91.512	5.111	0,756	15	*L. f.>C. g.*
Crural index	16	102.479	2.851	13	107.090	4.831	0,019	12	L. f.>C. g.
Ulna/radius index	16	116.173	1.017	22	114.320	1.768	0,003	15	*C. g.>L.f.**
Forelimb index	16	12.072	1.051	21	10.130	2.958	0,000	12	*C.g.>L.f.**
Hindlimb index	16	17.235	1.522	13	14.784	2.471	0,001	15	C.g.>L.f.*

*Significantly different after *t*-test (99%). Abbreviations: *C. g.* (*Callimico goeldi*), *L. f.* (*Leontocebus fuscicollis*).

Mature individuals of *C. goeldii* have longer hindlimbs relative to forelimbs compared to *L. fuscicollis*. In other words, at a given hindlimb length, *C. goeldii* has shorter forelimbs compared to *L. fuscicollis*, and these differences are maintained with age (Fig. 3). These results are supported by the least squares regression analyses (Table 2) as all regression slopes are under the isometric line, indicating that both primate species exhibit differential growth rates. However, the regression slopes also indicate that *C. goeldii* has a lower rate of growth compared to *L. fuscicollis*.

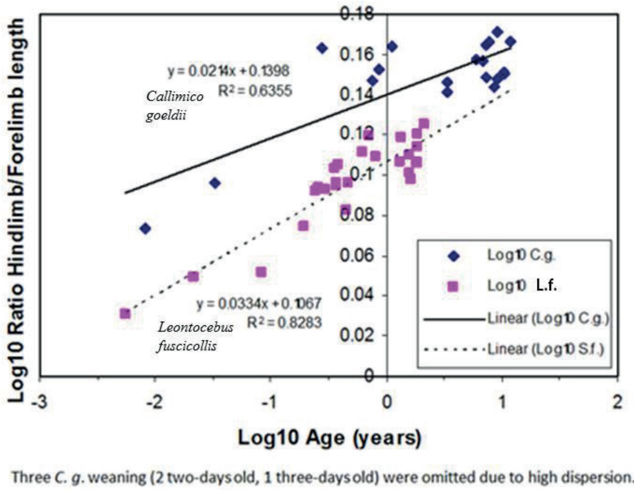

Fig. 3. Allometric comparison of the hindlimb/forelimb length ratio between *Callimico goeldii* and *Leontocebus fuscicollis*

Table 2. Least squares regression analysis between mature *Callimico goeldii* and *Leontocebus fuscicollis*

Primate	X axis (years)	Y axis (mm)	Intercept	Slope	R^2
Leontocebus fuscicollis	Age	HML	1.593	0.2372	0.876
	Age	RML	1.551	0.2411	0.870
	Age	UML	1.610	0.2486	0.875
	Age	FML	1.672	0.2729	0.915
	Age	TML	1.694	0.2692	0.901
Callimico goeldi	Age	HML	1.618	0.1530	0.948
	Age	RML	1.572	0.1533	0.937
	Age	UML	1.632	0.1615	0.944
	Age	FML	1.734	0.1698	0.920
	Age	TML	1.735	0.1805	0.944

All values in Log10

Piecewise regressions were performed on all immature and mature individuals, absolute *C. goeldi* postcranial maximum lengths were recorded in order to determine the age at growth cessation. Callimicos have an age at growth cessation of 14.9 months (Fig. 4). This age is similar to the one reported for sexual maturity in female callimicos (13–14 months, using radioimmunoassay of urinary steroid hormones), and the earliest recorded among all callitrichids (Dettling and Pryce 1999). Our *L. fuscicollis* data present two cluster groups that do not allow this type of regression. However, Garber and Leigh (1994), using captive-born saddle-back tamarin adult brain size data analyzed by a piecewise regression, reported growth cessation in the cranium at ~13.2 months. Fig. 5 shows scatter plots of maximum lengths *vs*. age with log-transformed trend lines indicating similar growth curves for both primate species.

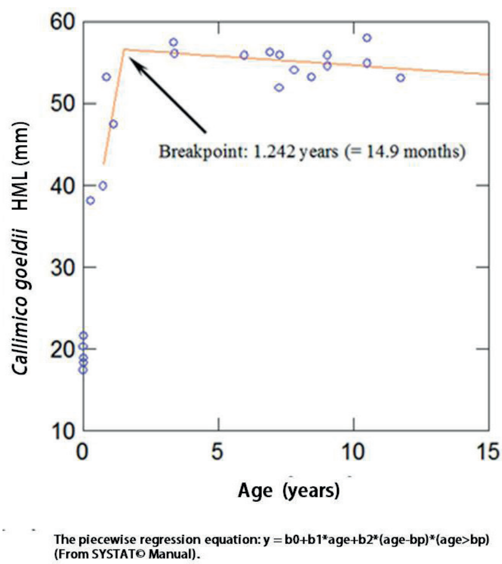

Fig. 4. *Callimico goeldii* humerus maximum length growth trajectory with the age of growth cessation

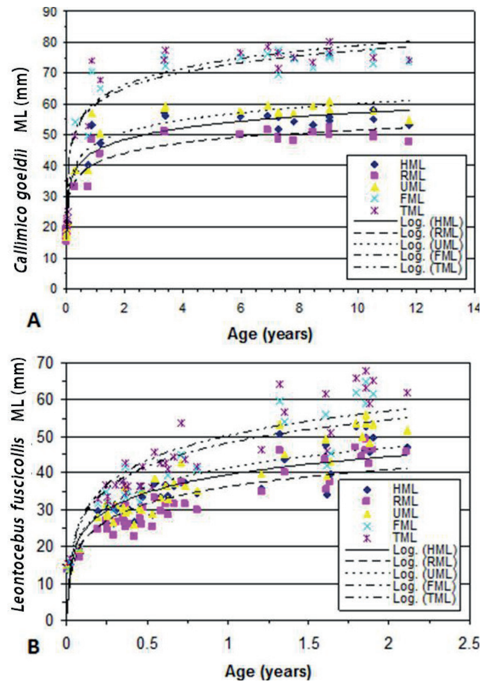

Fig. 5. *Callimico goeldii* and *Leontocebus fuscicollis* postcranial skeleton growth curves

Discussion

The results of the postcranial skeleton in *Callimico goeldii* and *Leontocebus fuscicollis* analyses provide insights into several aspects of their positional behavior. As indicated by Kimura (2003), arboreal primates tend to have longer hindlimbs than terrestrial ones, a feature related to the locomotor behavior adopted by each primate taxon in different environments. Garber and Leigh (2001a) argued that species differences in limb proportions rather than body mass offer a better explanation of differences in positional behavior and patterns of habitat utilization, which appears to be the case for other primate taxa as well (Garber 2007).

Growth trajectories showed in our study are congruent with those reported by Falsetti and Cole (1992), indicating that among callitrichids, saddle-back tamarins (*L. fuscicollis*), cotton-top tamarins (*S. oedipus*), and common marmosets (*Callithrix jacchus*), growth trajectories are also similar. In addition, as shown by Garber and Leigh (2001a) and Falsetti and Cole (1992), *L. fuscicollis* has proportionally longer forelimbs to hindlimbs during ontogeny compared to marmosets (*Callithrix* spp.) and other tamarins (*Leontocebus labiatus*).

Our results also show that, compared with *C. goeldii, L. fuscicollis* exhibits longer forelimbs than hindlimbs. Field studies on the positional behavior of these sympatric primate species suggest that longer forelimbs in *L. fuscicollis* may provide an "advantage by increasing the braking distance available for decelerating the body when landing in a rigid support" (Garber and Leigh 2001a: 28). In addition, in *L. fuscicollis* longer forelimbs might also be an adaptation for foraging, which is in accordance with data reported by Bicca-Marques et al. (1997, 1998) and Bicca-Marques (1999), who found that in this species, longer forelimbs might be an advantage during feeding.

On the other hand, in callimicos, elongated hindlimbs and a pattern of hindlimb development characterized by derived features of the ankle may serve to enhance stability during locomotion (Davis 1996). This has been argued to represent an adaptation for trunk-to-trunk leaping behavior (Garber and Leigh 2001a; Garber et al. 2009).

The differences in the postcranial skeleton and limb proportions in *L. fuscicollis* and *C. goeldii* suggest that different limb proportions, diet, foraging strategies, and patterns of habitat utilization enable these species to exploit different microenvironments in sympatry.

In sum, these data suggest that, in the evolutionary history of the Callitrichidae, differences in limb proportions and growth ontogeny might have played a major role in shaping ecological and behavioral differences between *C. goeldii* and *L. fuscicollis*. These differences include divergence in substrate use (Garber and Pruetz 1995; Heymann and Buchanan-Smith 2000; Berles et al. 2022), niche partitioning, feeding behavior (Bicca-Marques 1999), and positional behavior (Garber and Leigh 2001a, 2001b). It also indicates that limb proportions among callitrichids may be used to distinguish ecologically different taxa. Nevertheless, further research on the energetic cost of leaping, vertical clinging, quadrupedal running, and musculoskeletal design is needed (Warren and Crompton 1998; Polk 2002) in order to fully understand the specific relationships between limb morphology and positional behavior in the Callitrichidae.

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Conflict of interest

The author declared no conflict of interest.

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POLSKIE TOWARZYSTWO ANTROPOLOGICZNE

Spina Bifida Sacralis Occulta from Ancient Greek Sicily (Pozzanghera Necropolis, Leontinoi, 6th–4th Century BC): Anatomical, Anthropological and Ethnomedical Considerations on the Insular Presentation of this Congenital Anomaly

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Abstract: In this article a case of *spina bifida sacralis occulta* (SBSO) from the Pozzanghera necropolis of the ancient Greek colony of Leontinoi (Sicily) and archaeologically dated to the 6th–4th century BC is morphologically analysed and discussed. An ethnomedical excursus on the knowledge and impact of this condition in Sicilian history is offered. Although SBO is one of the most frequent congenital anomalies in the global palaeopathological record, a comprehensive literature search did not yield results for the history of Sicily before Christ, hence this case enriches existing knowledge on the historical presentation of this vertebral congenital anomaly in this region.

Key words: anatomy, congenital anomaly, palaeopathology, Sicily, *spina bifida occulta*, vertebral column.

Original article

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Introduction

The necropolis of the Pozzanghera district (*contrada*), located near Lentini (the ancient Greek town of Leontinoi, Λεοντῖνοι, home to the famed sophist Gorgias, 483–375 BC) in the present archaeological area in the province of Syracuse in eastern Sicily (Fig. 1), was subjected to systematic archaeological excavations between 26th October 1981 and 9th January 1982. The investigations were led by Prof. Dario Palermo (Full Professor of archaeology at the University of Catania and co-author of this article) and carried out in close collaboration with the Superintendency of Syracuse. The chronology of the site is very broad: the oldest tombs (*Tombs 37* and *38*) date to the Final Bronze Age (11th–9th centuries BC), therefore before the arrival of Greek settlers. Subsequently the Pozzanghera area was reutilised again as a necropolis starting from the 6th century BC (*Tomb 48*), while all the other burials date to a period comprised between the 5th and 4th centuries BC. At the end of the last excavation campaign 55 burials were brought to light (Palermo 1991). Among these, *Tomb 32* from Trench 7 (Fig. 2), which is the main focus of the present analysis, is bioarchaeologically relevant because of a vertebral anomaly detected in the skeleton found in this burial (Melintenda 2021), which will be discussed here.

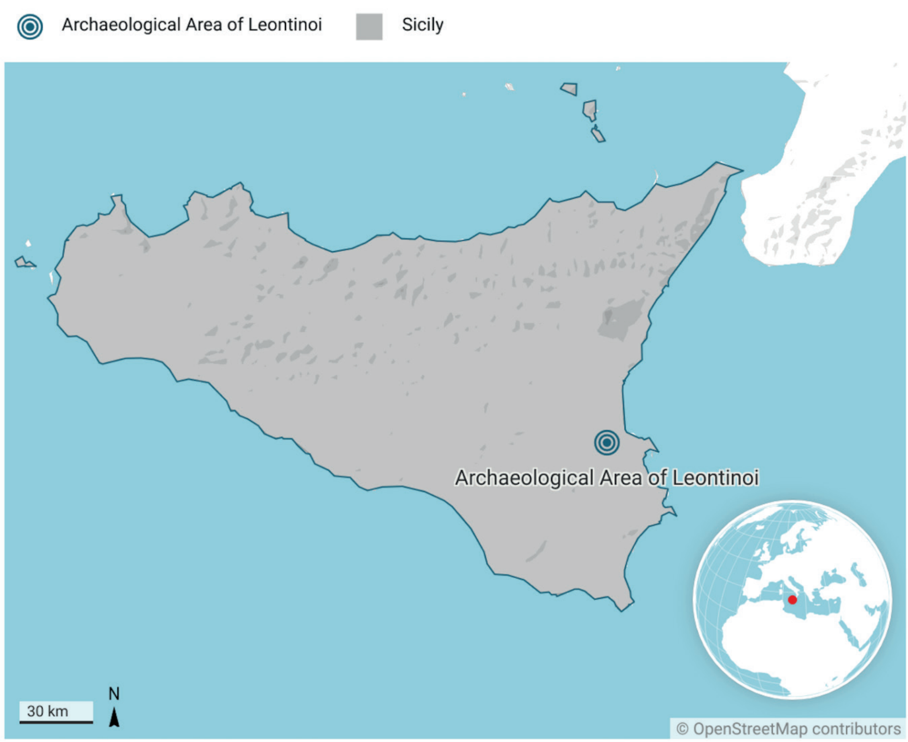

Fig. 1. Location of the Pozzanghera necropolis inside the Archaeological Area of Leontinoi

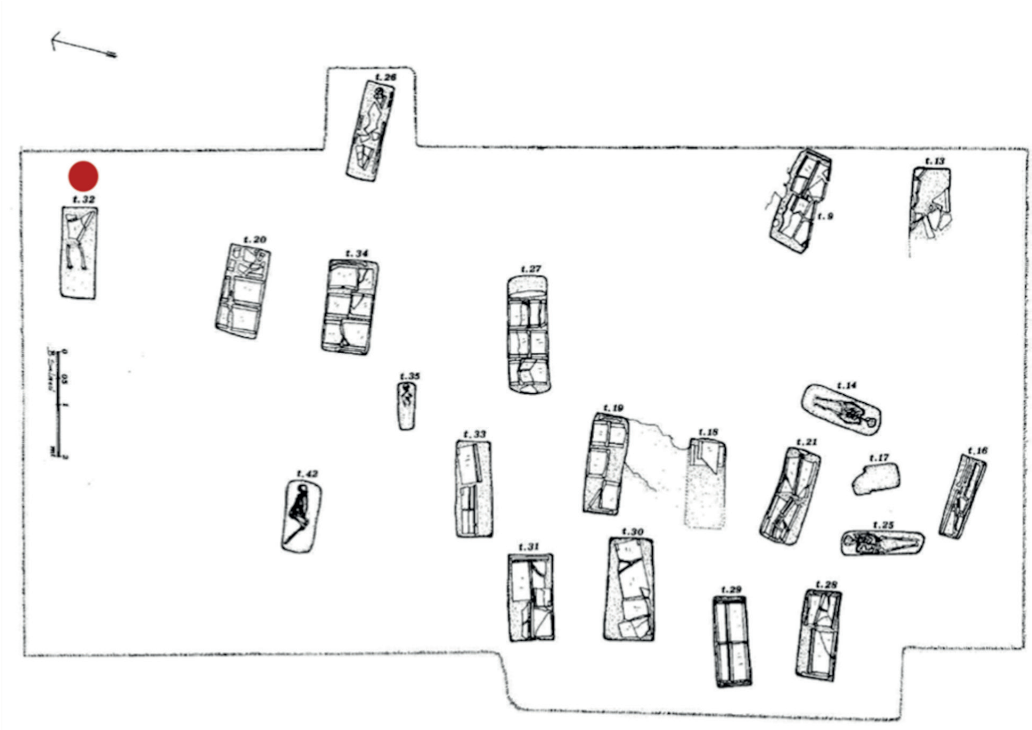

Fig. 2. Map of the Burials, Trench 7, from Palermo 1991. The red dot indicated the position of *Tomb 32*

Materials and Methods

The quantitative and qualitative analysis of the state of preservation were calculated by means of the Bello indices (2001): Bone Representation Index (B.R.I.) expressed as the ratio between the number of found bones and the theoretical number of bones that should be present in the actual total palaeodemography derived from the number of individuals retrieved in anatomical connection; Anatomical Preservation Index (A.P.I.) expressed as the ratio of conservation scores (in percentage) attributed to each bony element which constitutes the skeleton and the total number of bones of the skeleton (Bello 2001). The quality of the preservation of the cortical surfaces was evaluated through the Qualitative Bone Index (Q.B.I.), expressed as the ratio between the fully preserved cortical surfaces and the damaged ones for each skeletal segment (Bello 2001).

The sex of the individual was determined through the analysis of the morphological features of the retrieved fragments of the skull and pelvis (Ferembach et al. 1977–1979). Estimation of the individual's age at death was performed through the analysis of dental wear (Lovejoy 1985) and morphological changes of the auricular surface of the ilium (Lovejoy et al. 1985). Measurements of the sacral fragment's anomaly were taken using a digital sliding caliper.

The palaeopathological analysis was carried out by adopting a morphological, descriptive and metrical approach (Aufderheide and Rodríguez-Martín 2005; Ortner 2019; Varotto et al. 2021) and comparing the case with similar ones published in the specialised literature (Henneberg and Henneberg 1999). The sacral fragment was additionally subjected to radiological investigation. X-ray machine: Villa – Apollo EZ; X-ray parameters: 50kV 16mAs; CT scan machine: Siemens Somatom Sensation 16; CT scan parameters: 140kV 280mAs. A 3D reconstruction of the sacral fragment was performed using the OsiriX MD version 13.0.1 (Pixmeo).

Moreover, in order to chronologically contextualise the present palaeopathological report, a literature search was carried out on the free search engines *PubMed*, *Google Scholar*, *Wiley Online Library*, complemented by a *Google* and *Google Books* (English and Italian versions) search, using combinations of the following keywords: "spina bifida", "spina bifida occulta", "vertebral anomalies", "spine congenital disease", "neural tube defects" AND "Sicily", "Sicilian" AND "archaelogy", "anthropology", "bioarchaeology", "palaeopathology", "paleopathology", "archaeology", in order to identify publications that may help to reconstruct a chronological sequence of published reports of this condition in the bioarchaeological and palaeopathological record of Sicily. The main goal of this search was not making a comprehensive review of spina bifida in Sicily, but to verify if mentions of it existed in the specialised literature for the centuries before Christ, hence in a time frame including the chronology of Pozzanghera necropolis. In this search, non-indexed and non-bibliometric Italian-language archaeological sources were not included.

Results

At the time of its excavation, the skeleton found in *Tomb 32* lay in the supine position, with a bronze needle and a small 5th century BC λήκυθος /lékythos/ (a narrow-necked flask used in antiquity as a container for oils and ointments) next to the skull (Palermo 1991, Fig. 3). The preservation of the skeletal districts is partial. As far as the preservation indexes are concerned, they are as follows:

- B.R.I. = 35% (poor representation),
- A.P.I. = 50% (fairly good anatomical preservation),
- Q.B.I. = 47% (modest quality of the bone surface).

The individual's sex was determined to be male based on the morphologies of the external occipital protuberance and the mastoid processes at the cranial level and, in the pelvis, on the absence of the preauricular sulcus. As for the individual's age at the moment of death it was estimated to be of 24–30 years, hence a young adult based on dental wear, and 25–29 years (Phase 2) based on the morphology of the auricular surface of the ilium.

The sacrum is severely incomplete: only a small portion of it belonging to the first sacral vertebra and from the third to the fifth sacral vertebra have preserved to this day (Fig. 4). This more caudally preserved sacral segment is affected by an evident lack of closure of the neural arch at S3 – hence cranially to S4 where the sacral canal usually terminates with the sacral hiatus – to a presentation whose morphology is thus compatible with spina bifida occulta. Of note, this case of spina bifida is not only represented by the presence of the groove created by the lack of fusion of the posterior vertebral arches, but it also shows a considerable distance between the two edges of the neural arches.

The following measures of the patent sacral canal (S3–S5) are provided:

- Maximum length of the open sacral canal: 32.3 mm
- Maximum width of the open sacral canal: 19.5 mm.

These features are also palaeoradiologically observed (Fig. 5a–d), with the possibility, especially through CT transverse sections (Fig. 5b,c) to explore, in a cranio-caudal direction, how the posterior vertebral neural arch widens up to a complete separation creating the investigated dysraphism.

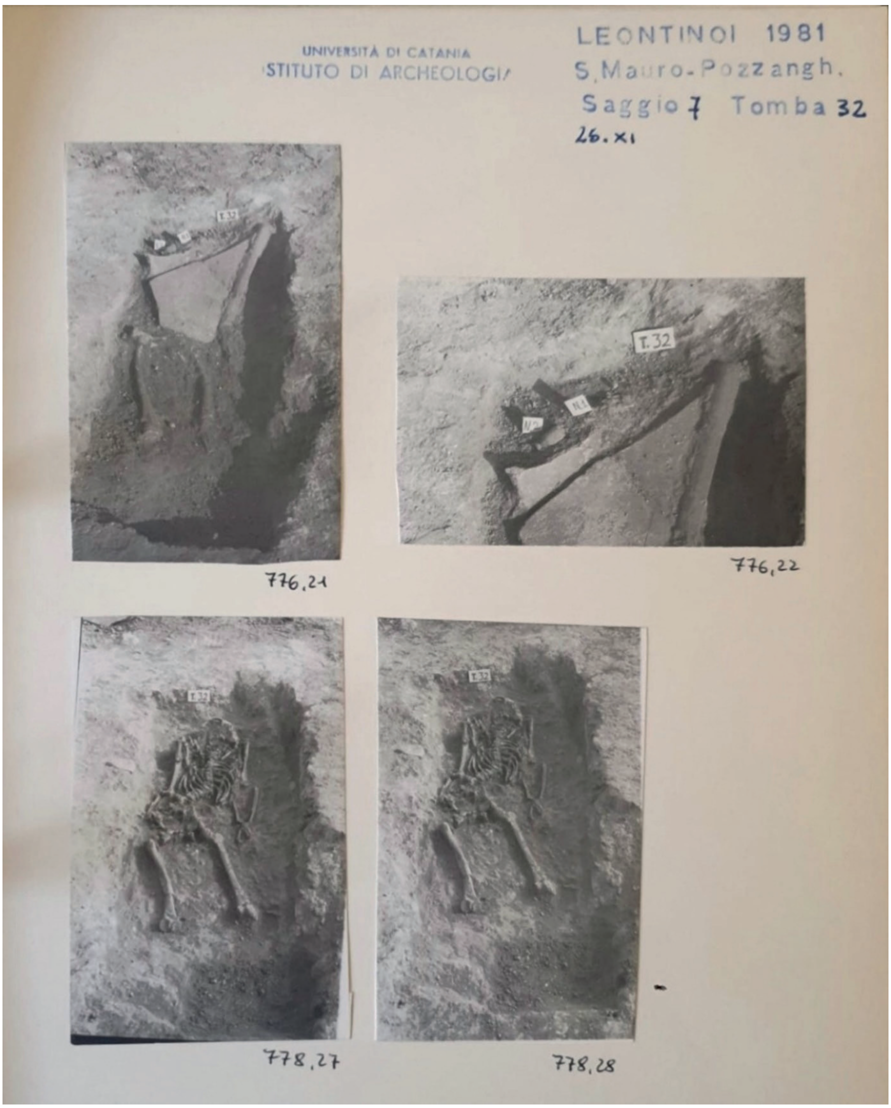

Fig. 3. *Tomb 32* at the moment of its complete excavation. Photos from Dario Palermo's personal archive

Fig. 4. Dorsal view of the fragment of the sacrum from *Tomb 32* with *spina bifida sacralis occulta*

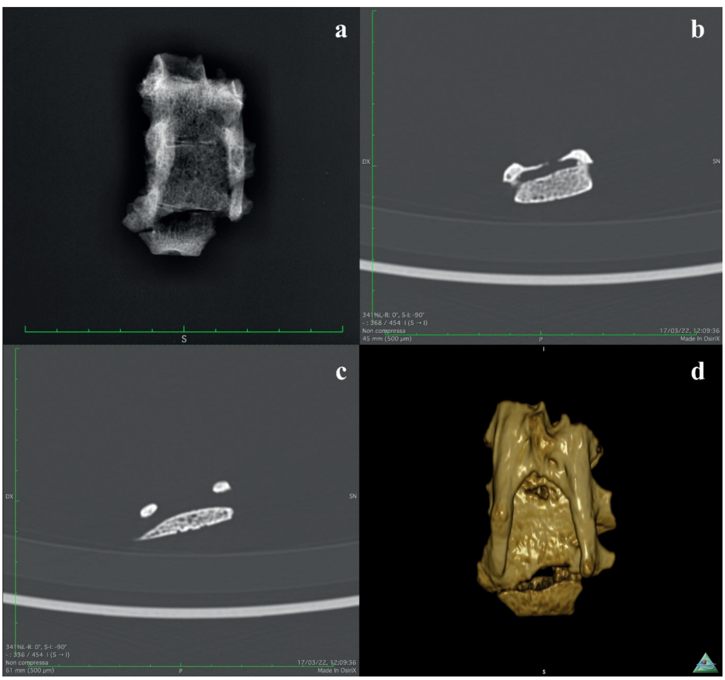

Fig. 5.a. X-ray image of the sacral fragment in postero-anterior projection
Fig. 5.b. CT scan image (transverse section) at the origin of the dysraphism (L3)
Fig. 5.c. CT scan image (transverse section) at a more caudal level (L4), well within the sacral hiatus
Fig. 5.d. 3D virtual reconstruction of the sacral fragment with spina bifida in dorsal view

Discussion

This condition occurs when an incomplete bone line appears on one or more neural arches. It is found in the lumbosacral region, most frequently in the sacrum (*spina bifida sacralis*). Spina bifida can occur in two different variants, occulta (SBO) and aperta (SBA) (Aufderheide and Rodríguez-Martín 1998). SBO consists of an incomplete fusion of the posterior neural arch and may affect one or more sacral segments. SBO can involve one or two vertebrae: in particular, the first sacral vertebra is statistically the most affected one, as highlighted by studies on living patients such as the review of 1,200 radiological images of Israeli patients which showed that 17% of them had that specific anatomical location (S1) (Nastoulis et al. 2019). The meninges do not protrude outside of this defect. Thus, spina bifida occulta can be diagnosed radiologically in living patients, often not earlier than late childhood or adulthood as it may well lead to no relevant disabilities. Nonetheless, in the number of reported clinical consequences of SBO the cutaneous stigma of hypertrichosis as well as disc pathology, genitourinary dysfunction, intraspinal lipoma, etc. are counted (Gregerson 1997).

On the contrary, SBA is a much more severe condition than SBO, and can lead to fatal outcomes. It is linked with various pathologies, such as hypoplasia of the vertebrae, diastematomyelia (split cord malformation), scoliosis, kyphosis and hemivertebrae.

There are three degrees of severity (Aufderheide and Rodríguez-Martín 1998):

- *Meningocele*: meninges and nerve roots extrude through the neural arch defect, but the spinal cord remains *in situ*. The lumbosacral region is the most commonly affected site and accounts for 5% of cases of SBA.
- *Myelomeningocele*: the spinal cord is also extruded. This type constitutes the majority of cases of SBA (about 60%) and affects the L-S segment. It may be associated with hydrocephalus.
- *Myelocele*: in this case the skin does not close at the level of the vertebral anomaly, hence causing the newborn to succumb shortly after birth as a result of infectious processes. This is the most serious type of SBA and can result in death from infection.

Furthermore, it is worth underlining that for palaeopathologists spina bifida represents a problematic matter because it is difficult to distinguish between its two types: skeletal remains are generically referred to as spina bifida occulta (SBO), since it is not possible to know what external presentation of the anomaly, at the cutaneous level, actually was in life. According to Morse the presence of a meningocele or myelomeningocele causes severe neurological symptoms and is incompatible with a long life, which leads to the conclusion that, whenever an archaeologist finds this defect in an adult skeleton, it is safe to consider the condition as spina bifida occulta (Morse 1978). To this definition Barnes's can also be added: "The spinal canal is widened with neural tube defect, pushing the edges of the bony cleft outward. In contrast, the spinal canal remains normal, and the edges of the bony cleft are not raised when no neural tube defect is present" (Barnes 1994). The former definition fits SBA, whereas the latter is more adequate for SBO.

From an historical perspective, among the early descriptions of spina bifida feature that by the ancient Greek physician

Hippocrates (ca. 460-ca. 375 BC) in the ancient world and by the Dutch clinician Peter van Forest (1522–1597) and the Swiss scholar Caspar Bauhin (1560–1624) in the Early Modern Period (Goodrich 2008; Ashwal 2021). However, its first illustrated description and its very scientific name are to be owed to the work of Nicolaes Tulp (1593–1674) – at a time in which anthropological research did not exist as a separate scientific branch and was still part of the greater family of human anatomy (Papa et al. 2019) –, the famous Dutch anatomist immortalised by Rembrandt's 1632 painting *De anatomische les van Dr Nicolaes Tulp* ("The Anatomy Lesson of Dr. Nicolaes Tulp") exhibited in the Mauritshuis art museum (The Hague, the Netherlands), in his work *Observationes Medicae* in 1641 (Goodrich 2008). Tulp reported on some cases of spina bifida and used this definition as chapter XXX of this work (*Spina dorsi bifida*, "Bifid spine of the dorsum"), where he described, with full anatomical arguments, a case of spina bifida aperta (*vidimus spinae medullam plane laceram; et nervorum propagines tam varie, per tumorem dispersas*, "we saw the spinal medulla completely torn and the nerves' endings scattered in such varied degrees", Fig. 6) (Tulp 1641), which resulted in the infant's death due to what would now be defined an incorrect surgical procedure, namely the ligation and surgical removal of the herniated neural contents.

Fig. 6. Illustration of a case of spina bifida aperta from Nicolaes Tulp's 1641 work, book III, page 235

However, its first complete description is to be attributed to the German pathologist Friedrich Daniel von Recklinghausen (1833–1910), who in 1886 published his work *Untersuchungen über die Spina bifida* (Goodrich 2008) – without forgetting the contributions to the study of this anomaly by the Sicilian physician Giuseppe Muscatello (1866–1951) (Tardieu et al. 2017). Furthermore, Rudolf Virchow (1821–1902), while commenting on the case of a Greek soldier with sacral trichosis (Fig. 7) reported on by Dr Bernhard Ornstein, chief physician of the Greek Army, presented a case he himself had examined, that of a 24-year-old female (Fig. 7): *Als ich nunmehr den Rücken präparierte, stellte sich sonderbarerweise heraus, dass unmittelbar unter der behaarten Stelle eine Spina bifida occulta lag, d. h. dass an dieser Stelle die Wirbel an ihrem hinteren Umfange nicht geschlossen waren. An der Stelle der Dornfortsatze der oberen Kreuzbeinwirbel befand sich eine harte Membran; nachdem dieselbe eingeschnitten war, sah ich* *unmittelbar die Dura mater spinalis vor mir*. ("When I now dissected the dorsum, it turned out strangely that immediately below the hairy part there was a spina bifida occulta, i.e. that at this point the vertebrae were not closed at their posterior circumference. In the place of the spinous processes of the upper sacral vertebrae there was a hard membrane; after it had been cut into, I immediately saw the spinal dura mater in front of me" (Ornstein & Virchow 1875).

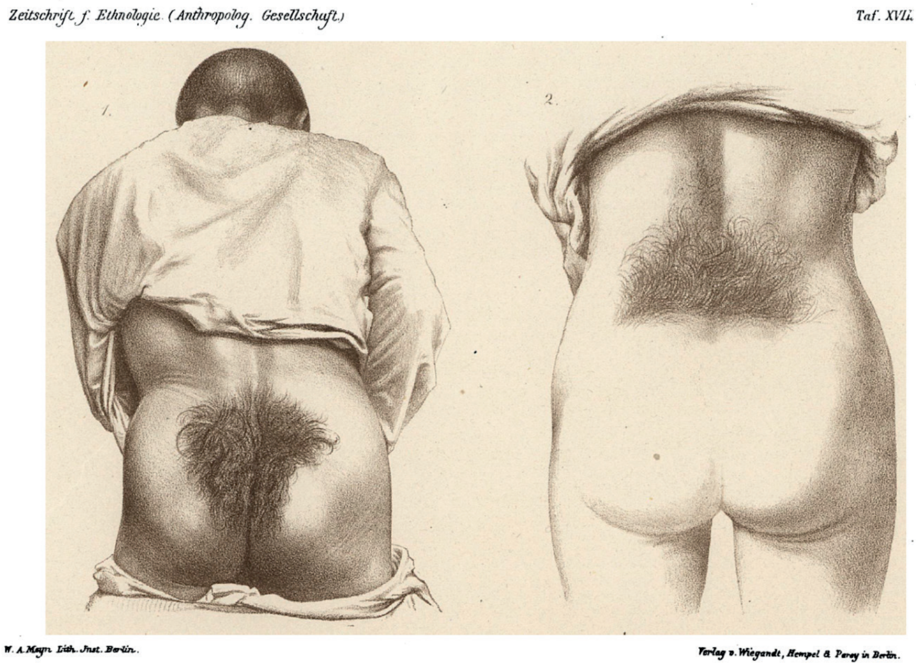

Fig. 7. Illustration of cases of spina bifida occulta in Rudolf Virchow's work from *Zeitschrift für Ethnologie* 1875, 7. Bd., Table XXVII, Figs. 1 and 2

With particular reference to SBO, it is described as one of the commonest congenital anomalies affecting the spinal column (Kelty and Henneberg 2022). A recent review indicated a statistically significant increase of this condition after AD 1900 and linked this increase in frequency with relaxed natural selection (Kelty and Henneberg 2022), namely the result of environmental elimination or weakening of specific sources of selection that proved formerly key to the preservation of a particular trait (Staub et al. 2018).

In the case from Leontinoi, while a standard morphological assessment permitted a *prima facie* diagnosis, unlike in most studies mentioning this anomaly, the implementation of palaeoradiological techniques was stressed. In *in-vivo* studies on patients in the contemporary clinical setting, MRI and ultrasound imaging are the standard diagnostic techniques, while CT scan imaging has been described as of limited value in the spina bifida *in vivo* because of its low sensitivy and poor tissue constract, with the further burdern of exposure to iodising radiation for the patients (Trapp et al. 2021). It is generally solely utilised in cases where it proves impossible to sedate a patient or the investigated bone anomaly is severe (Trapp et al. 2021). However, it happens more frequently that following an X-ray or CT scan imaging session for diagnostic purposes, spina bifida can be an incidental report (Manenti et al. 2016). Nonetheless, it should be underlined how CT scan imaging of mummied human remains can actually yield precise information on the presence of spina bifida (Picchi et al. 2022). In the presently studied case, since no patients nor soft tissues were involved, the implementation of X-ray and CT scan imaging helped to better understand the underlying architecture of this congenital anomaly.

Based on these morphological and radiological considerations it can thus be concluded that the described *spina bifida sacralis* is of the occult type, considering that SBA would be incompatible with the age at death estimated for the individual from *Tomb 32*.

Additional palaeopathological information on this skeleton's spine includes the possibility that it may also have been affected by a severe form of scoliosis, formulated at the time of the excavation by observing the *in-situ* curvature of spine (Palermo 1991), but this notion is to be probably ruled out after an examination of the remnants of the vertebral column and ribs in the laboratory.

As for the exact cause of this vertebral anomaly, it is impossible to identify it retrospectively, since, only having one single line of evidence – in this instance osteological in nature –, and no pathobiographical information on this individual *intra vitam* nor genetic data, it is preferable to restrain oneself from venturing into the field of aetiology (Galassi and Gelsi 2015).

Excluding causes which are not possible for ancient times (intake of neural tube defect-causing medications such as valproic acid, an antiepileptic) or are less likely (obesity and diabetes, rarer risk factors than they are in the modern world and clearly not showing their non-specific lesions on the discussed skeleton), other aetiologies such as folate deficiency, hyperthermia in the early weeks of pregnancy, or family history of neural tube defects can be carefully considered (Mitchell et al. 2004). As to folate (vitamin B9) deficiency, its role in neural tube defects (NTDs) was unknown until the 1960s when the British paediatrician Richard Worthington Smithells (1924–2002) and obstetric pathologist Elizabeth Hibbard (1928–2019) noted the association (Wald 2011), which ultimately led scientists in the early 1990s to establish that an increased intake of folate or folic acid in the early phases of pregnancy reduces the risk of NTDs (Bentley et al. 2006). As a result, in 1998 the US Food and Drug Administration made it mandatory that folic acid fortified enriched grain products (Bentley et al. 2006), whereas, still in the late 1990s in Sicily, women at risk for recurring neural tube defect were reported in a study not to be aware of the prevention of such outcomes through folic acid supplements (Pepe et al. 1999). While the role of folates in the aetiology of spina bifida was not well-known or understood at the popular level in the Sicilian population, it is important to underline how, on the contrary, the morphological presentation of this neural tube defect was indeed known. The Sicilian physician and ethnologist Giuseppe Pitrè (1841–1916), in his 1896 work *Medicina po-*

polare siciliana ("Sicilian Popular Medicine"), reported that spina bifida was called in the Sicilian language "la cusuzza" (standard Italian: "la cosuccia"; English: "the little thing") and defined it as follows: *È un tessuto incancrenito, e se si tocca disattentamente si corre pericolo di far morire il bambino. Si strofina con saliva per tre giorni di seguito; e sparisce al terzo* ("It is an encrusted tissue, and if you touch it carelessly you run the risk of killing the child. You rub it with saliva for three days in a row; it disappears on the third day") (Pitrè 1896). Pitrè's description seems to refer to the aperta variant of spina bifida in that a protruding mass would account for the word "cusuzza" and the quasi-magical attempts at treating it he described.

Regarding the role of dietary folates in the pathophysiology of this condition, a recent study by Mutlu and colleagues (2020) showed how a Mediterranean Byzantine population which had access to folate-rich food products presented a lower prevalence of spina bifida than other Anatolian populations taken as a comparison.

As for the antiquity of the reported cases, the extensive literature search performed did not yield any reports of this condition in Sicily for the centuries before Christ. This gap in the literature should not lead to an underestimation of the prevalence of this vertebral anomaly in the past as it could be explained by the fact that it may have gone unnoticed by past archaeologists with no to minimal training and/or practical experience in osteoarchaeology, or may simply have not been reported in scientific or multidisciplinary publications due to its being considered unimportant if weighted against other more purely archaeological aspects.

Nonetheless, even though more reports were or will be available in the future, as noted by Henneberg and Henneberg (1999), «[d]espite many published reports on spina bifida occulta, from both historical and modern skeletal collections, it is difficult to compare frequencies of spina bifida occulta across populations and through time», due to the underlined «lack of a clear definition of spina bifida occulta in the sacrum for use in comparative skeletal studies».

Conclusions

This report of a Greek-era Sicilian case of *spina bifida sacralis occulta* helps to expand knowledge on the presence of this condition in the ancient history of the Sicily. Future reports of this condition might lead to the possibility of an assessment of the prevalence of this anomaly in the midst of the Mediterranean region.

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Conflict of interest

The authors declare no competing interests.

Authors' contribution

Co-first authors S.M. and E.V. wrote the first draft with E.V. acting as co-supervisor of the thesis of S.M. and having a major role in the methodological planning. E.P., L.G., V.P., D.P. all contributed with archaeological or review remarks, revised the original draft and offered substantial criticism that was incorporated in this manuscript. F.M.G supervised the work, wrote the discussion, contributed to the palaeoradiological analysis together with E.V. and V.P.

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Tooth size discrepancies in Kosovar adolescents with different malocclusion classes

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Abstract: The goal of this study was to evaluate tooth size discrepancies in Kosovar adolescents according to the Bolton's analysis, and to determine the differences between gender and malocclusion classes.

A sample of 400 Kosovar adolescents, aged 13–19 years with various malocclusion categories (class I, II, III) according to the Angle's classification, was randomly selected. The anterior tooth size ratio, overall tooth size ratio, posterior tooth size ratio, as well as distribution of tooth size discrepancies were assessed. The normality of distribution was assessed by the Kolmogorov-Smirnov test, the differences between genders by the Independent Sample T-test, the Mann-Whitney U-test, and differences among malocclusion groups by ANOVA, the Kruskal-Wallis, and the Dunn's post-hoc tests.

The tooth size ratios of men and women did not differ significantly. The results also demonstrated significant differences among the malocclusion classes only for the anterior tooth size ratio ($p<0.05$). The overall and posterior ratios did not differ significantly within malocclusion classes. The percentages of Kosovar subjects with a deviation of more than 2SD from the Bolton average for the anterior and overall ratios were 41.37 and 23.79, respectively.

The genders did not differ significantly regarding tooth size ratios. Among the malocclusion classes, the anterior ratio differed significantly. In Kosovar subjects, tooth size discrepancies tend to be higher compared to Bolton's averages.

Key words: dental cast analysis, tooth size discrepancies, Kosovar adolescents, Bolton analysis.

Original article

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Introduction

Tooth size discrepancies are commonly characterized as a significant redundancy of dental tissues in one dental arch compared to the opposite side (Fields 1981) but also as a disparity in the sizes of specific teeth (Proffit 2007). Although natural teeth are well matched in most people, Proffit (2007) estimates show that around 5% of the population has tooth size disparities.

To diagnose and treat orthodontic problems correctly, it is important to determine whether there are tooth size disparities between the upper and lower jaws. As a result, orthodontic treatment plans are altered by decreasing (interproximal reduction), enlarging (crowns or build-ups), or removing the teeth before completion (Cançado et al. 2017).

Orthodontists have used various methods to identify discrepancies between dental arches in patients. However, the Bolton analysis, based on the ratios of the mandibular and maxillary mesiodistal tooth diameters, is the most well-known and popular technique (Smith and Buschang 2000).

According to Bolton (1958), the mesiodistal tooth size of the maxilla and mandible should correspond to the best occlusion, overjet, and overbite at the end of orthodontic treatment. However, an intermaxillary tooth size disparity is among the numerous indicators that can impair the success of orthodontic treatments.

The interrelationship between tooth dimension ratios and malocclusion groups has been studied in the last century until today, and different results have been obtained. Currently, we have no information about tooth size discrepancies in our population. Consequently, the objective of this study was to identify tooth size discrepancies and to determine the significance of differences between genders and malocclusion classes among Kosovar adolescents.

Material and methods

Study population

The sample of this study included 400 teenagers (216 females and 184 males) aged 13 to 19 years (average age 15.17 years ±1.91 SD). Participants were residents of seven regional cities of the Republic of Kosovo and were randomly selected by the multistage cluster sampling in 14 different schools. The study's inclusion criteria were as follows: Kosovar nationality, aged 13–19 years; fully erupted permanent teeth, except third molars; no previous or ongoing orthodontic treatment; no tooth abrasion, attrition, or major restorations that might have affected a tooth's mesiodistal dimension; no fractured teeth, no abnormal tooth morphology; and good quality study casts. According to the Angle classification (1899), the 400 study participants were classified into three categories: Class I (N = 212), Class II (N = 166), and Class III (N = 22) (Figure 1). This study was approved by the Ethics Committee of the School of Dental Medicine at the University of Zagreb (05-PA-30-XXIII-1/2021).

Methods

An informative letter and a consent form were given to all study participants who fulfilled the criteria and were signed by them (if they were over 18 years old) or by their parent/witness (if they were under 18 years old).

Fig. 1. The distribution of the sample according to gender and malocclusion groups (n=400)

Alginate was used to take impressions of the maxilla and mandible, and pre-orthodontic casts were made. Direct measurements of the MD width of teeth, from first molar to first molar, were taken of the pre-orthodontic casts with an electronic digital caliper (CD-6''ASX; Mitutoyo Corp., Kanagawa, Japan) to an accuracy of 0.01 mm (Figure 2).

Fig. 2. Measurement of the tooth size (MD width)

Bolton analysis

Tooth size ratios were determined following the Bolton's analysis (1958; 1962). The Bolton anterior ratio was calculated by dividing the sum of the widths of the lower and upper frontal teeth (from the canine to the canine) of both jaws. The result was then multiplied by 100.

$$\textit{anterior ratio} = \frac{\sum (33 \leftrightarrow 43)}{\sum (13 \leftrightarrow 23)} \text{ x100}$$

The Bolton overall ratio was calculated by dividing the sum of the widths of the lower and upper teeth (from the first molar to the first molar) of both jaws. The result was then multiplied by 100.

$$\textit{overall ratio} = \frac{\sum (36 \leftrightarrow 46)}{\sum (16 \leftrightarrow 26)} \text{ x100}$$

The Bolton posterior ratio was calculated by diving the sum of the widths of the lower and upper posterior teeth (from the first premolar to the first molar, on both sides of the jaws). The result was then multiplied by 100.

$$posterior\ ratio = \frac{\sum(36\leftrightarrow 34,44\leftrightarrow 46)}{\sum(16\leftrightarrow 14,24\leftrightarrow 26)} x100$$

All measurements were performed by the same researcher (B.Z.L.). To assess the examiner's measurement error, 30 randomly selected pairs of casts were premeasured 24 hours later by the same investigator. An Independent Sample T-test was used to determine the measurement error. The Dahlberg's formula was used to determine the deviation, ranging from -0.72 to 0.47 mm (Dahlberg 1940) (Table 1).

Each malocclusion type's percentage of tooth size disparity was calculated using the Bolton's standard. The entire number of study participants in the group was divided by the number of participants with tooth size ratios greater than 2SD. This value was multiplied by 100 (Strujić et al. 2009) (Figures 3 and 4).

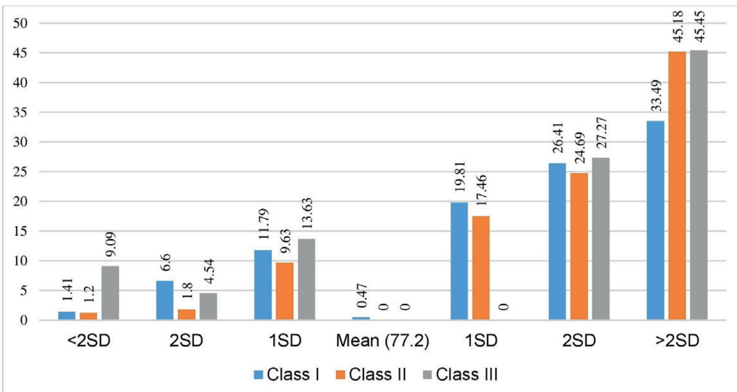

Fig. 3. The proportion of 400 participants with an anterior tooth size discrepancy compared to Bolton's norms

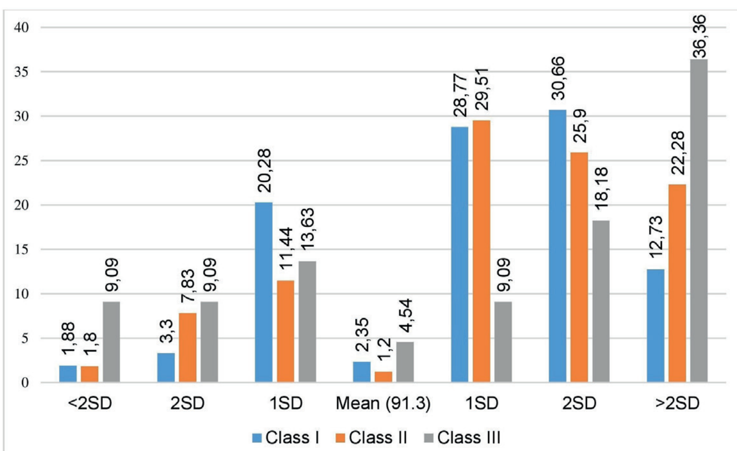

Fig. 4. The proportion of 400 participants with an overall tooth size discrepancy compared to Bolton's norms

Statistical analysis

Statistical analysis of the data was performed using SPPS version 25.0 (New York, USA).

Normal data distribution was analyzed using the Kolmogorov-Smirnov test. The data for the anterior tooth ratio were not distributed normally, while the data for overall and posterior ratios were distributed normally. Therefore, males and females were compared using the Independent Sample T-test and Mann-Whitney U-test. The variation among the three classes of malocclusions was analyzed using ANOVA, Kruskal-Wallis, and Dunn's post-hoc tests. A statistically significant p-value of 0.05 was used.

Results

The results of the T-test indicated that the differences between the two groups of measurements were not statistically significant (Table 1).

The descriptive data for the anterior, overall, and posterior ratios are shown in Table 2. Males and females did not show statistically significant differences ($p > 0.05$).

The Kruskal-Wallis test revealed that the anterior ratio differed significantly between malocclusion classes ($p<0.05$) (Table 3). Moreover, in subjects within class II, the greatest values for anterior ratio ($p<0.05$) were found. The ANOVA test revealed no significant differences in overall and posterior ratios between genders and malocclusion groups (Table 3).

On the other hand, the Dunn's post-hoc test showed significant differences in anterior ratio, more specifically between classes I and II ($p<0.05$) (Table 4).

Table 1. Descriptive statistics of tooth size ratios (n=30)

	Mean		SD		Mean difference	Lower	Upper	P-value	Dahlberg's error
	M1	M2	M1	M2					
Anterior ratio	79.93	79.46	2.81	2.82	0.47	-0.98	1.93	0.51	0.47
Overall ratio	92.46	92.48	2.32	2.25	-0.19	-1.37	0.99	0.74	-0.19
Posterior ratio	105.47	106.20	3.54	3.50	-0.72	-2.54	1.09	0.42	-0.72

Table 2. The differences in tooth size ratios between genders (n=400)

	Female and Male		Female		Male		F	Mean difference	P-values†,‡
	Mean	SD	Mean	SD	Mean	SD			
Anterior ratio ‡	79.81^	2.95	79.83^	3.06	79.80^	2.83	0.28	0.04	0.71
Overall ratio	92.89	2.62	92.84	2.73	92.97	2.50	0.39	-0.13	0.63
Posterior ratio	105.99	3.74	105.79	3.76	106.23	3.73	0.06	-0.44	0.24

Abbreviations: SD-Standard deviation; VAR-Variance; †Independent Sample T-test; ‡ Mann Whitney U-test; ^ Median

Table 3. The mean, standard deviation (SD), Kruskal-Wallis, and analysis of variance (ANOVA) tests for the anterior, posterior, and overall ratios and anterior and overall discrepancies in different malocclusion groups (n=400)

	Anterior ratio		Overall ratio		Posterior ratio	
	Mean	SD	Mean	SD	Mean	SD
Class I	79.37	2.69	92.69	2.27	106.02	3.22
Class II	80.31	2.87	93.12	2.77	105.96	4.22
Class III	80.41	4.90	93.21	4.20	105.97	4.78
P-value ‡	**0.012****		0.232		0.922	
FEMALE	Anterior ratio		Overall ratio		Posterior ratio	
	Mean	SD	Mean	SD	Mean	STD
Class I	79.30	2.76	92.45	2.23	105.59	3.11
Class II	80.30	2.71	93.21	2.81	106.01	4.24
Class III	80.92	5.69	93.45	4.72	105.89	5.12
P-value *†*	**0.024***		0.093		0.735	
MALE	Anterior ratio		Overall ratio		Posterior ratio	
	Mean	SD	Mean	SD	Mean	SD
Class I	79.44	2.62	92.95	2.29	106.48	3.28
Class II	80.33	3.08	93.01	2.73	105.91	4.23
Class III	79.31	2.50	92.69	3.05	106.12	4.35
P-value	0.108		0.947		0.607	

*, ** Statistically significant at 0.05; SD-Standard deviation; † ANOVA test; ‡ Kruskal-Wallis test

Table 4. Multiple comparisons between malocclusion classes in anterior ratio by post-hoc Dunn's test

Pairwise Comparisons of Classes					
Sample 1-Sample 2	Test Statistic	Std. Error	Std. Test Statistic	Sig.	**Adj. Sig.**[a]
I-III	-30.069	25.896	-1.161	.246	.737
I-II	-34.791	11.982	-2.904	.004	**.011**
III-II	4.722	26.231	.180	.857	1.000

Each row tests the null hypothesis that the Sample 1 and Sample 2 distributions are the same.
Asymptotic significances (2-sided tests) are displayed. The significance level is .050.
a. Significance values have been adjusted by the Bonferroni correction for multiple tests.

Discussion

Tooth size discrepancies have been investigated all over the world to compare populations with specific features and identify trends in variability among dental arches. Their significance in orthodontic diagnosis is well documented in scientifically publications. The orthodontic society has acknowledged the importance of the interrelationship between the maxillary and mandibular teeth to complete treatment (Araujo et al. 2003).

According to the current research, the average anterior tooth size ratio was 79.81 percent, which was higher compared to the Bolton's study (Table 2). Moreover, both genders and all malocclusion classes showed an average overall tooth size ratio of 92.89 percent, which was also higher compared to the value of 91.3 percent in the Bolton study (Table 2). The differences between populations and samples in the two studies can, to some extent, explain these findings. For example, Bolton used a small homogeneous group (55 Caucasian females) with excellent occlusion, whereas our study was conducted on schoolchildren and included 400 non-orthodontic subjects of both genders and different malocclusions.

Tooth size discrepancies and their prevalence

In 1962, Bolton suggested that a deviation from the average of over 1 SD indicates a requirement for diagnostic attention. His study found that 29% of his private practice patients had tooth-size disparities exceeding one standard deviation (Bolton 1962). In contrast, other researchers (Crosby and Alexander 1989; Freeman et al. 1996) interpreted the requirement as more than 2SD deviations from the Bolton standard. According to this, several studies have defined the prevalence of tooth size disparities and reported different results.

In the current study, the frequency of a significant disparity (more than 2 SD) in the anterior ratio was 41.37 percent (Figure 3), which is in accordance with previous findings in other populations (Akyalçin et al. 2006; Wedrychowska-Szulc et al. 2010; O'Mahony et al. 2011). Higher values indicate a trend for the mandibular tooth to be oversized in participants with class III. This suggests that the anterior maxillary teeth were smaller in subjects with class III compared to class II and class I. According to Akyalçin et al. (2006), there may have been considerable individual and cultural diversity in the growth pattern among the respondents.

In contrast, percentage values for anterior discrepancy ratio have been reported in Dominican American (Santoro et al. 2000), Southern Chinese (Ta et al. 2001), Brazilian (Araujo et al. 2003), Japanese (Endo et al. 2008), Jordanian (Al-Omari et al. 2008), Croatian (Strujić et al. 2009), Turkish (Uysal et al. 2005; Oktay and Ulukaya 2010), American (Johe et al. 2010), and Libyan (Bugaighis et al. 2015).

The incidence of a significant overall ratio discrepancy in the current study was 23.79 percent (Figure 4), which was similar to the results reported in a study carried out in a Turkish population (Oktay and Ulukaya 2010).

In contrast, Bolton (1958; 1962) and Proffit (2007) observed under 5 % of individuals with an overall ratio disparity of more than 2 SD. However, their studies comprised individuals with perfect occlusion, which could be assumed to be more typical of the normal community than of orthodontic patients. The prevalence obtained in the current study, however, was higher than reported in previous studies (Bolton 1958, 1962; Santoro et al. 2000; Bernabé et al. 2004; Proffit 2007; Al-Omari et al. 2008; Endo et al. 2008; Strujić et al. 2009; Wedrychowska-Szulc et al. 2010; Oktay and Ulukaya 2010; Bugaighis et al. 2015). However, Akyalçin et al. (2006) reported greater prevalence value compared to our study. Their study sample was drawn from an orthodontic population, which could explain why they had the highest percentage of anterior tooth size discrepancies.

The higher frequency of statistically significant anterior tooth size discrepancies in the Kosovar population compared to the overall discrepancies suggests a significantly larger number of participants with proximal anterior tooth size disparities exhibiting more than 2 SD from the Bolton mean compared to study participants with overall disparities. The reason for this might be that the frontal teeth, particularly the upper and lower teeth, are significantly more prone to tooth size deviations. In other words, the anterior region exhibits the highest variability in mesiodistal tooth sizes (Uysal et al. 2005; Oktay and Ulukaya 2010). The latter might indicate that the prevalence of the Bolton discrepancy may differ between populations with different occlusal disorders. Therefore, clinicians ought to be aware of the frequent occurrence of TSDs while assessing and treating orthodontic patients. As a result, regardless of the malocclusion group, gender, or population, conducting Bolton's study routinely is strongly encouraged (Strujić et al. 2009; Johe et al. 2010).

Tooth size ratios and gender

Various studies have reported gender differences in tooth size proportions. In our study, no substantial difference was found between men and women according to the Bolton's tooth size ratios (Table 2). Previous studies (Crosby and Alexander 1989; Nie et al. 1999; Santoro et al. 2000; Ta et al. 2001; Araujo et al. 2003; Bernabé et al. 2004; Basaran et al. 2006; Al-Omari et al. 2008; Endo et al. 2008; O'Mahony et al. 2011; Bugaighis et al. 2015; Ismail and Abuaffan 2015; Mujagic et al. 2016; Hashim et al. 2017; Machado et al. 2018) have also reported no gender differences in tooth size proportions. On the other hand, some previous studies (Moorrees et al. 1957; Lavelle 1972; Richardson and Malhotra 1975; Smith et al. 2000; Uysal et al. 2005) compared the tooth size ratios between men and women and found considerable disparities. For example, Moorrees et al. (1957) found gender differences only in the overall ratio. Lavelle (1972) compared the overall and anterior tooth ratios between males and females and concluded that males had the highest ratios. Smith et al. (2000) discovered that men had considerably higher overall and posterior ratios compared to women. Oktay and Ulukaya (2009) noted sexual dimorphism only for the posterior ratio. In contrast, Richardson and Malhotra (1975) reported no changes in tooth-size proportions between the anterior and posterior arches. Mollabashi et al. (2019) discovered a substantial gender disparity in the posterior and overall ratios, whereas Strujić et al. (2009) found gender differences only in the anterior ratio. Therefore, the above studies suggest that a variation related to tooth size ratios depending on gender must be calculated separately for each population. Overall, most studies reveal no significant difference between tooth size ratios and gender.

Tooth size ratios in different malocclusion classes

The fact that tooth size variation is not systemic proves that populations differ in terms of tooth size ratios between arches. The variation in maxillary tooth size by population and gender is not correlated with differences in mandibular tooth size, so that different tooth size ratios are observed between arches (Smith et al. 2000).

The current study showed that only the anterior ratio differed significantly among the malocclusion classes ($p<0.05$), but neither the overall ratio nor the posterior ratio differed significantly (Table 3, 4). On the other hand, Strujić et al. (2009) and Oktay and Ulukaya (2009) reported contrasting findings. There were no significant differences in the anterior tooth ratios among the malocclusion classes. They did, however, discover substantial changes in the overall and posterior ratios. Moreover, several previous studies (Ta et al. 2001; Uysal et al. 2005; Basaran et al. 2006; Al-Khateeb et al. 2006; Lopatiene and Dumbravaite, 2009; O'Mahony et al. 2011) among different populations reported no significant variation between the malocclusion classes in anterior and overall ratios. Other researchers (Sperry et al. 1977; Nie et al. 1999), on the other hand, reported that the overall tooth size ratios were higher in class III than in classes I and II. According to Araujo and Souki (2003), class III subjects had a substantially larger anterior tooth size discrepancy than class I and II subjects. The tendency toward greater tooth size proportions in class III was also observed in the Chinese population by Ta et al. (2001).

Comparison between gender and classes

The present study found no substantial disparities between the genders and classes in the anterior, overall, and posterior ratios (a p-value of 0.05) (Table 3). The results of our study are consistent with other studies (Smith and Buschang 2000; Akyalçin et al. 2006; Endo et al. 2008; Alam et al. 2013; Cançado et al. 2015), which showed no significant gender differences in Bolton ratios in the anterior and overall regions by malocclusion category. On the other hand, according to Lavelle (1972), tooth size varies across different occlusal categories and populations. The results of our study were not consistent with those reported by Fattahi et al. (2006), who discovered gender variation in the anterior ratios of malocclusion classes but not in the overall ratios.

Further, O'Mahony et al. (2011) reported that males in classes II division 2 and III had the largest average anterior tooth size ratios, as opposed to classes I and II division 1. In contrast, Uysal and Sari (2005) noted no significant differences between the genders within the normal occlusion group, except for the overall ratio in the Turkish population.

Strengths and limitations

Although our study is not without its limitations, we recognize that it has some strengths as well. One of the strengths of our study is that the schoolchildren participating in the study were sampled from different cities, indicating a representative sample. In addition, they all met the study requirements. On the other hand, one of the limitations can be that our sample consisted of a general population with different malocclusions. Another limitation is that the measurements were made using a 2D method rather than a 3D method. Therefore, future work should focus on orthodontic Kosovar subjects and use a 3D method for the measurements.

These results suggest that the national criteria for clinical status are required. As the Bolton statistics are not representative of the non-orthodontic Kosovar population, these data should not be used in regular orthodontic diagnosis and treatment for Kosovar orthodontic patients. As a result, clinicians need to be aware of the occurrence of TSDs and therefore, Bolton's analysis is necessary.

Conclusions

The following conclusions can be derived from our study's findings:

1. The averages and standard deviations of the anterior and the overall ratios were higher in the current study than in Bolton's research.
2. The tooth size ratios of men and women did not differ significantly.
3. Class II malocclusion showed the anterior ratio difference between classes.
4. Among Kosovar adolescents with more than two SD from Bolton's averages, the anterior tooth size ratio was 41.37 %, and the overall tooth size ratio was 23.79%.

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Conflict of interests

None.

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None.

Authors' contributions

BZL: Conceptualization, Methodology, Validation, Writing-Original draft preparation, reviewing, and editing. BK: Data curation, Visualization. SM: Writing-Reviewing and Editing, Supervision, Project administration.

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Assessment of dental caries among children and adults inhabiting Starorypin (11^{th}–12^{th} c.) compared to other Early Medieval populations from Poland

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ABSTRACT: Dental caries testing provides information about diet, hygiene and eating habits. This information can be useful in reconstructing the living conditions of historical populations. The aim of this study was to determine the frequency of dental caries in adults and children in the Early Middle Ages population from Starorypin (Poland) and compare the results with other early medieval populations from Poland.

A total of 346 adult permanent teeth and 131 deciduous teeth were included in the study. Both the macroscopic method and imaging methods (X-ray and a light-induced fluorescence technique) were applied in the study.

Dental caries were recorded in 33% (115/346) of the permanent teeth and in 10% (13/131) of the deciduous teeth. In permanent teeth, dental caries were diagnosed on the approximal surface, while dental caries in deciduous teeth are most often diagnosed on the occlusal surface. Early childhood caries (ECC) were mainly diagnosed on the teeth of individuals within the 3.0–5.5 years age group.

The frequency of dental caries seemed to be high compared to other early medieval populations. The obtained results may have been influenced by 'broadband' odontological research methods, not limited to the macroscopic method alone. However, it can be also argued that the frequency of dental caries observed in this study might have been influenced by a highly varied diet, as indicated by archaeozoological and archaeobotanical studies. The surfaces affected by dental caries correspond to findings of other studies showing that in adults, these were the approximal surfaces, while they were the labial/occlusion surfaces on the deciduous teeth.

KEY WORDS: dental caries, Starorypin, Early Medieval.

Original article

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Introduction

Nowadays, the assessment of the condition of the dentition is a common aspect of bioarchaeological research (Ioannou and Henneberg 2017; Koruyucu and Erdal 2021). This is due to the fact that the odontological material is well preserved at archaeological sites, even where bone materials are subject to significant fragmentation and destruction.

One of the diseases assessed in bioarchaeological materials is dental caries (Yanko et al. 2017; Moles 2023). Based on the analysis of the frequency of dental caries, eating habits and/or the level of hygiene can be assessed indirectly. However, it must be remembered that oral hygiene and eating habits are only one of the cariogenic factors. Clinical practice indicates that other causative factors may be congenital hypomineralization of enamel, malocclusion, tooth displacement in alveolar arch or the chemical composition of saliva (Fejerskov and Kidd 2003). However, in the historical materials, not all of the indicated factors are possible to determine.

Additionally, the results of odontological research can be interpreted in the context of archaeobotanical, archaeozoological or isotopic research (Tomczyk et al. 2020a; Schata et al. 2022). This information is crucial for understanding the economic and social status of studied populations. Therefore, in the odontological studies of historical populations, caries studies play an important role.

In this work, attention will be focused on the period of the early Middle Ages in Poland. In the period up to 1138, in the lands between the Oder and the Bug Rivers a centralized state ruled by the Piast dynasty was established, which were known as 'Poland' from the 10th and 11th centuries (Urbańczyk 2008). Although quite a lot of early medieval burial grounds in Poland have been reported (e.g., Cracow, Wroclaw, Milicz) (Miszkiewicz and Gronkiewicz 1986; Kwiatkowska 2005; Mietlińska 2015), ‚broadband' odontological examinations have been only carried out in selected ones (e.g., Cedynia, Ląd, Brześć Kujawski, Radom) (Kozubkiewicz et al. 1957; Kozubkiewicz and Trachtenberg 1960; Borysewicz and Otocki 1975; Borysewicz-Lewicka and Otocki 1978). However, a lot of these studies dated back to the 1960s and 1970s. This means that each new site from the early Middle Ages examined in terms of odontology is valuable for understanding this period in the history of Poland.

The aim of this study was to assess changes in the diet and level of hygiene among young children (the age of 1–8 years) and adults who inhabited Starorypin (Poland) in the early medieval period, and compare the studied population with other selected populations from early medieval Poland. This study aims to address the following questions: i) what was the frequency of dental caries in children and adults, ii) where were dental caries most often located, iii) in what age groups among children were caries most common and iv) how does the studied group present itself against groups from other sites of the early Middle Ages?

Materials and Methods

Archaeological context

The Starorypin site is located in North Eastern Poland, approximately 65 km from Toruń (Fig.1). Studies of the settlement complex in Starorypin indicate that it was an important settlement in the early and late Middle Ages and an administrative center of the interfluve of the Vistula, Drwęca and Skrwa. The key to research on the history and culture of the early medieval Dobrzyń land has been the discovery and partial recognition of the cemetery from this period (site 6). The location of the necropolis remained unrecognized until recently and the work was focused on former central places and settlements (Lewandowska 2016, 2022).

Fig. 1. Location map of the study area

The history of research on early medieval burials in Starorypin dates back to the 1960s. At that time, following heavy rains, human bones used to be observed in the escarpment. As a result of the archaeological intervention, six very poorly preserved skeletons were recorded (Grześkowiak 1963, 1967). Another accidental unveiling of a fragment of the necropolis took place in 2010, when human remains were found on the heap during a linear investment. As a result of excavations (until 2022) and anthropological analysis, 52 graves and 29 objects classified as bone clusters were recorded. Within the excavated graves, 68 individuals were identified, while 29 individuals were identified in the group of loose bones.

The fragment of the necropolis examined has provided, apart from human bones in various states of preservation, a representative historical material, constituting an equipment and personal belongings of the deceased, indicating various fields of production and material manifestations of everyday life and spiritual culture. In particular, numerous grave inventories have been discovered in the burials of adult women. The largest group of all items have been ornaments, primarily necklaces made of glass beads and other materials, including semi-precious stones (carnelian, amethyst, rock crystal) and amber. The findings also included temple rings and other rings. Following ornaments, the second largest group of items have been tools and everyday objects.

The observed funeral rite (orientation of graves, skeletal burial) can be associated with Christian eschatology. On the surface of the necropolis examined to date, the confirmed form of the grave was a flat grave (i.e., a grave devoid of any superstructures visible on the surface). The basic feature of the layout of early medieval skeletal cemeteries is the row arrangement of graves, which is also observed at the Starorypin site (Bojarski 2020; Stawska 2003; Stawska et al. 2010).

Based on the typologies of objects deposited in the graves and the analysis of the obtained radiocarbon dates, fragments of ceramic vessels and reflections on the functioning of the entire settlement complex, including the stronghold and the suburban settlement, it was possible to determine the time of functioning of the cemetery, which was operating from the first half of the 11th century to the second half of that century and into the 12th (Błędowski and Lewandowska 2022).

Dental materials

A total of 25 adults (males: 12, females: 13), with a total of 346 permanent teeth (males: 181, females: 165), and 14 children, with a total of 131 deciduous teeth were examined.

Regarding adults, the basic criterion for material selection was the possibility of establishing the sex and age-at-death of the individuals. The sex and age-at-death of individuals were assessed according to the commonly accepted anthropological methods (Brooks and Suchey 1990; Buikstra and Ubelaker 1994).

The examined children were divided into three age groups: 1.0–2.5 years, 3.0–5.5 years and 6.0–8.0 years. These age groups reflect the successive stages of tooth eruption (Liversidge and Molleson 2004). Because among children no permanent teeth have been preserved, only the deciduous dentition was used.

The diagnosis of dental caries was carried out through visual, radiographic and fluorescent techniques:

(i) Visual observation of the dental caries was conducted using a 3x dental magnifying glass and a sharp dental probe. Visual inspection was performed under a direct dental unit light.

(ii) We used radiographic techniques for diagnosis; for this task, a portable X-ray machine (EZX-60, Edlen Imaging, USA) was used.

(iii) A light-induced fluorescence technique was used for the detection of initial enamel caries on occlusal and approximal surfaces. For this task, we used the VistaCam iX Proof device (Dürr Dental, Germany).

The location of carious lesions (approximal, root and occlusal) was considered and analyzed. When more than one surface was affected, each surface was analyzed separately.

In the deciduous dental analyses, particular attention was paid to early childhood caries (ECC). This kind of caries appears in caries-resistant regions, such as the labial surfaces of the upper incisors, the upper and lower molars and, more rarely, in the upper canine and (even more rarely) in the lower canine and incisors (Çolak 2013; Begzati et al. 2015).

Fig. 2. Examples of the advancement of caries in adults (A: No 61(1)/2022, B: No 61/2022)

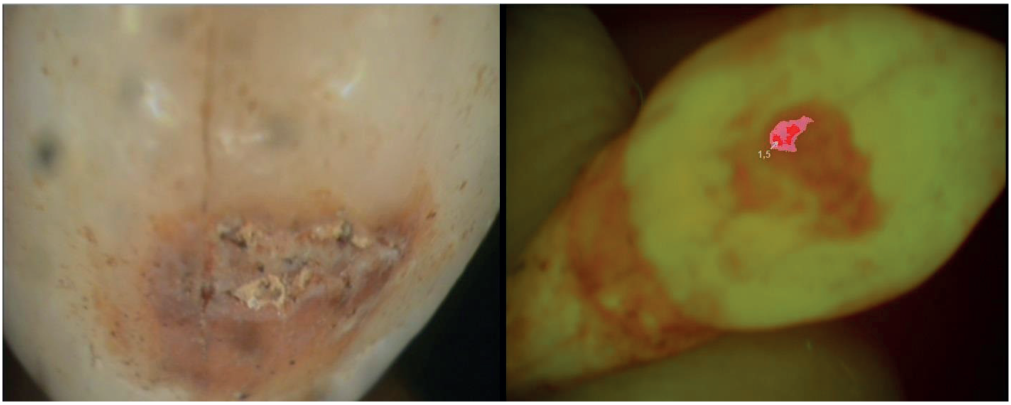

Fig. 3. Caries on the labial surface of the deciduous canine (FDI: 83) macroscopic photo and using fluorescent light (No: 35/2020)

The frequency of caries (i.e., the number of teeth affected by caries) was calculated based on the proportional correction factor (PCF) following Erdal and Duyar (1999, 2003). The PCF was applied because dental materials are often missing in historical materials (e.g., during post-mortem deposition). Ofter, the anterior teeth are missing due to their morphological structure. The PCF provides caries rates of anterior and posterior teeth according to their appropriate numbers: i) permanent teeth: three-eighths for anterior and five-eighths for posterior teeth; ii) deciduous teeth: three-fifths for anterior and two-fifths for posterior teeth, multiplied by the number of caries observed.

The analyses also included an assessment of the severity of carious lesions. Carious lesions were divided into (i) enamel decay (early-stage tooth decay, which is formed before a cavity) and (ii) cavities.

In order to estimate the intra-observation and inter-observation error, 80 teeth were chosen at random. These analyses were performed by two experienced odontologists (JT, DO-K).

The chi-squared test was used to analyze the differences in caries frequency. In addition, due to the small samples, the Fisher's Test was used. The chi-squared test was also used to compare measurements in the intra- and inter-observation error estimations. Statistical analyses were performed using the R software (http://www.R-project.org. 2013). Differences with $p < 0.05$ were considered statistically significant.

Results

The intra- and inter-observation error was found to be statistically insignificant ($p = 0.6732$ and $p = 0.6223$, respectively).

Out of 25 adults, 20 were diagnosed with dental caries, while caries were observed in 4 of the 14 children. Dental caries were recorded in 33% (115/346) of the permanent teeth and in 10% (13/131) of the deciduous teeth. Dental caries were found in 26% (43/165) of females and 40% (72/181) of males; however, the observed differences are not statistically significant ($p = 0.0912$).

Dental caries in adults were diagnosed most frequently on the approximal surfaces and least often on the root. In contrast, in children, dental caries were most often diagnosed on the occlusal surface. In both cases, the observed differences were statistically significant.

Analyzing children in successive age categories, a clear increase in dental caries in the second age category (3.0–5.5) was observed. Changes in dental caries frequency between age categories were statistically significant (Fisher Test: $p = 0.0056$, Monte-Carlo simulations: $p = 0.0040$). Although there were not many tested individuals, ECC was diagnosed on all deciduous teeth of children aged 3.0–5.5 years (Table 1).

Table 1. Frequency of dental caries in the population from Starorypin

	Total	anterior	posterior	ECC#	approximal	root	occlusal
Adult	115/346 (33%)	2/115 (0.65)*	113/115 (61.4)*	-	79/115 (69%)	27/115 (23%)	46/115 (40%)
					$p < 0.0001$ $\chi^2 = 48.852$		
Children	13/131 (10%)	4/13 (18.4)*	9/13 (27.6)*	13/13 (100%)	6/13 (46%)	0/13 (-)	11/13 (85%)
					$p < 0.0001$ $\chi^2 = 18.979$		
1.0-2.5	1/58 (2%)	0/1 (-)	1/1 (40)*	1/1 (100%)	1/1 (100%)	0/1 (-)	0/1 (-)
3.0-5.5	12/61 (20%)	2/12 (10)*	10/12 (33)*	12/12 (100%)	6/12 (50%)	0/12 (-)	10/12 (83%)
					$p = 0.0002$ $\chi^2 = 17.1$		
6.0-8.0	0/12 (-)	0/0 (-)	0/0 (-)	0/0 (-)	0/0 (-)	0/0 (-)	0/0 (-)

*proportional correction factor (PCF).
early childhood caries (ECC).

Table 2. Frequency of caries in selected sites from the early Middle Ages in Poland

Archaeological site	Period	Frequency of teeth with dental caries	References
Kraków-Zakrzówek	11th–13th c.	3.1%	Gleń 1975
Brześć Kujawski	11th c.	9.8%	Kozubkiewicz et al. 1957
Kałdus	11th–12th c.	10.4%	Kozubkiewicz and Trachtenberg 1960
Garbary	12th–13th c.	12.0%	Borysewicz-Lewicka and Otocki 1978
Cedynia	11th–13th c.	13.1%	Stopa and Perzyna 1978
Ląd	11th–13th c	14.0%	Borysewicz and Otocki 1975
Stary Brześć	12th–16th c.	14.0%	Borysewicz and Otocki 1975
Ołbin	12th–13th c.	14%	Kwiatkowska 2005
Wrocław	12th–13th c.	15.7%	Staniowski et al. 2011
Wrocław – pl. Dominikanski	12th–14th c.	22%	Kwiatkowska 2005
Radom	11th–12th c.	38.0%	Tomczyk et al. 2020b
Starorypin	11th–12th c.	33.0%	presented research

In this study, an attention was paid to the degree of advancement of dental caries, separating enamel decay from cavities. On the permanent teeth, 23 (20%) enamel changes and 96 (83%) cavities were observed, while on the deciduous teeth, 8 (62%) cases of enamel decay and 5 (38%) cavities were observed.

Discussion

Compared to data obtained from other archaeological sites from the early Middle Ages in Poland, the occurrence of dental caries in Starorypin is considerably higher (Table 2). This may be due to a small sample size, although it cannot be ruled out that the low frequency of teeth with caries results from a different methodology, e.g. an examination based only on macroscopic assessment, without the use of a fluorescent camera or X-ray. Lucas et al. (2010) showed that the use of the radiological method increases the detection of caries. Also, Tomczyk et al. (2014), showed that the use of micro-CT and VistaCam iX Proof increases the detection of early dental caries. Comparative studies have shown that these methods of dental caries detection were not used, which could to some extent underestimate the detection of the disease, especially in its early stages. This supposition may be supported by the comparability of the results from Starorypin with another Polish early medieval population, such as the Radom population, which was studied using the same devices and a similar methodology (Tomczyk et al. 2020b). In the early medieval population from Radom, for example, dental caries were identified in 38% of permanent teeth.

The assessment of the frequency of dental caries is used in archaeological research to reconstruct living conditions, health and/or hygiene habits of past populations (Yanko et al. 2017; Moles 2023). Although the frequency of teeth infected with caries seems to be significantly high at the Starorypin site, it should be remembered that this level was clearly higher in subsequent historical periods. The high frequency of teeth infected with caries observed at at the Starorypin site has been explained by a monotypic economy and a cariogenic diet (O'Sullivan et al. 1993; Kurek et al. 2009; Bertilsson et al. 2022).

The reconstruction of the diet of the Starorypin population was made on the basis of archaeozoological and archaeobotanical analyses. This research showed a diverse picture of the environment, which was probably resulted in a high diversity of the produced foods. For example, it was shown that the local population not only raised mammals (cattle, pigs, sheep) and poultry but was also engaged in hunting, as evidenced by the presence of deer, roe deer and wild boars and fishing (pike, carp and tench) (Makowiecki 2022). Fertile areas were dominated by deciduous forests with a predominance of hornbeam and poor sandy sites that were covered with pine and pine-oak forests. On the other hand, wetlands around water reservoirs and swamps were occupied by communities dominated by alder. Lands used to be acquired not only for crops and pastures, but also for housing estates and roads (Noryśkiewicz 2022). The conducted analyses show that diet of the early medieval population was not limited to cariogenic products, such as cereals. Instead, this population exhibited a rather varied diet containing a large proportion of animal protein. High-content protein diets have been shown to reduce the acidity of the saliva and, thus, neutralize the decalcifying acids that cariogenic bacteria produce. The consumption of diets that are relatively high in protein increases blood urea levels, which may lead to relatively high salivary urea levels. Since urea appears to be the main substrate for the formation of dental plaque, a slight increase in the salivary urea concentration or output may reduce the development of caries (Dawes 1970; Silverstone et al. 1981; Zabokova-Bilbilova et al. 2012). However, research on diet composition in the above studies requires further isotope studies to confirm this interpretation.

In this study, attention was focused on the location of dental caries. In the studied population, dental caries dominated on the approximal surfaces. This can be explained by intensive chewing, conditioned by the type of food, which potentially led to a strong abrasion of the occlusal surfaces. This deprived the teeth of numerous retention places where food could linger, destroying the crown of the tooth, while the poor level of hygiene meant that the remains of the food were in the cervical regions long enough to undergo fermentation and start the process of enamel destruction. Similar observations apply to other early medieval populations from Poland, such as Cedynia, Słaboszewo and Góra Chełmska (Borysewicz and Otocki 1975; Stopa and Perzyna 1978; Torlińska-Walkowiak and Jerszyńska 2011).

The examination of the degree of dental caries advancement showed that carious lesions were diagnosed in the majority. Given that the age of most individuals at the time of consent was not high (20/35 years old) (Myszka et al. 2022), it can be assumed that dental caries had either an acute course or developed at a very early age. Unfortunately, observations of the severity of caries were carried out sporadically in populations from the early Middle Ages. However, a similar observation was reported on the early medieval population from Chełmska Góra (Borysewicz and Otocki 1975) and Garbary (Borysewicz-Lewicka and Otocki 1978). Interestingly, in these studies enamel decay was most frequently observed among children. It can be assumed that the development of these changes was halted by the death of the individual.

In the analyzed population of Starorypin, children with deciduous dentition were also included. Overall, dental caries were diagnosed in 10% of children. Due to the scarcity of archeological studies on children, it is rather difficult to compare the results of our study to other early medieval sites in Poland. In Garbary (12–13th c.), dental caries were diagnosed on the deciduous teeth of children in the *infans I* category at the level of 3% (Borysewicz-Lewicka and Otocki 1978). Equally, low dental caries on deciduous teeth were recorded on the primary teeth of children from Chełmska Góra (Borysewicz and Otocki 1975). In the *infans I* group from Cedynia, the total frequency on the tooth type level of carious lesions was 2.8% (Torlińska-Walkowiak and Jerszyńska 2011). However, the cited studies did not divide the tooth material into three age categories, but, instead, analyzed the tooth material as one age group of *infans I*. This certainly does not facilitate comparative analyses.

Dental caries in the deciduous dentition allows for inferences on weaning and feeding practices that promote dental decay. For instance, a lower frequency of carious lesions suggests a simpler diet based on low in cariogenic products. The Starorypin population may have had more access to processed and sweetened foodstuffs and, therefore, refined carbohydrates were more readily available. In our study dental caries were most often observed among children aged 3.0–5.5 years. This would suggest that the process of introducing solid products started before the age of three, which corresponds with population isotope studies from the early Middle Ages in Radom showing that the weaning period occurred roughly between 1.1 and 2.5 years of age (Tomczyk et al. 2021). However, the presented interpretation must be confirmed by isotope studies.

The dental caries among children were diagnosed on the occlusal surfaces, which, due to a slight abrasion, exhibited many furrows, providing places for the accumulation of food remains. Similar observation regarding the location of caries on deciduous teeth was reported in the Góra Chełmska site (Borysewicz and Otocki 1975).

When examining the teeth of nonadults, it is worth paying attention to early childhood caries (ECC) (Çolak 2013; Begzati et al. 2015). This kind of dental caries is also known as „severe early childhood caries" or „nursing caries". Newly erupted teeth do not have fully matured enamel; it is thinner, with large dental tubules. In addition, compared to adults, salivation is slower in children (Rohnbogner and Lewis 2016) and decay processes develop quicklier. Therefore, the development of ECC is characterized by highly dynamic changes, which, in a short time, lead to the destruction of the tooth crown and, consequently, to the development of diseases of pulp and periapical tissues. It is generally accepted that ECC develops by the age of 5 years. The dynamics of changes in ECC are impossible to detect in bioarcheological materials because it is not possible to determine the rate of decay. However, their location can be helpful for identification. This kind of caries appears in caries-resistant regions: labial surfaces of the upper incisors, in the upper and lower molars and, more rarely, in the upper canine and (even more rarely) in the lower canine and incisors (Çolak 2013; Begzati et al. 2015). Although the material in our study is sparse, ECC can be observed primarily among individuals aged 3.0–5.5, although ECC was also observed in the earlier period (1.0–2.5 years).

Conclusion

Our examination of the masticatory apparatus, narrowed down to the assessment of dental caries, is an important element of bioarchaeological research. The paper presents analyses performed on the early medieval population discovered in Starorypin. Although the discovered burials are not numerous and require further isotope analyses, they broaden our knowledge about the early medieval population of Poland. Furthermore, the study investigated not only the permanent dentition of adults, but also the deciduous dentition of children.

The presented analyses show that the population of Starorypin probably functioned on a varied diet, which did not generate pro-inflammatory processes. The location of dental caries on the approximal surfaces in adults and on the occlusal surfaces in children corresponds to the pattern which was also observed in other populations from this period.

Conflict of interests

The Authors do not have any conflict of interest.

Authors' contribution

AM: assessment of the sex and age of the individuals; DO-K: dental analysis, preparation and description of the manuscript; MZ: statistical analysis; WN: statistical analysis; JL: archaeological and historical information; JT: dental analysis, planning and supervision of the research, setting a goal, substantive supervision.

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Anthropometric characteristics of four groups of indigenous people of Wallacea in East Indonesia

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Abstract: The human body size varies significantly around the world, both between and within populations. Likewise, ethnic diversity has a significant effect on anthropometric data differences for the Naulu, Tana Ai, Bugis and Rampasasa tribes, in Wallacea, East Indonesia. Six body dimensions were collected from 484 people, 219 males and 265 females in the age from 18 to 80 years. The statistical analysis included tabulating the means and standard deviations for the various body dimensions and proportions. A one-way ANOVA with post hoc LSD test was performed to determine significant differences between the means of anthropometric dimensions and proportions, as well as within the four ethnic groups. There are significant differences ($p<0.01$) among the four ethnic groups and two sexes in most measurements taken. The post-hoc LSD test indicated that the Naulu male has the largest body size compared to the Tana Ai, Bugis, and Rampasasa tribes. The Naulu have a large body size, a long trunk, broad shoulders, and long legs. Tana Ai and Bugis people have nearly identical characteristics, namely a medium body, long trunk, narrow shoulders and hips, whereas Rampasasa people have a small body size, with a long trunk, narrow shoulders and a wide pelvis. The Naulu and Bugis people have trapezoidal trunks, while the Tana Ai and Rampasasa people have rectangular trunks. In conclusion, the tribes in the Wallace area of East Indonesia have a wide variety of physical characteristics. Further research is needed to understand how changes in technology, development, transportation and large migration flows affect the demographic and physical characteristics of ethnic groups in Indonesia.

Key words: anthropometry, ethnicity, Wallacea, body dimensions.

Original article

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Introduction

All around the world, humans have striking variations in body sizes. In terms of an evolutionary and biogeographical context, the variation in human body size results from the worldwide distribution of human populations, exposure to multiple environments, evolutionary forces, and from complex forms of cultural behavior (Little 2020). The population of indigenous people is estimated to be only about 400 million people worldwide, but represents an extraordinary cultural diversity (Funk et al. 2020). As a very diverse country, Indonesia has different ethnic groups, who can be classified as indigenous peoples. The population census by the Central Statistics Agency in 2010 revealed that Indonesia has around 1,300 ethnic groups, as reported in the official website of the Ministry of State Secretariat (Central Statistics Agency 2021). These ethnic groups not only show differences in daily customs, but also have distinctive morphological characteristics.

There are many ethnic groups living in Eastern Indonesia, particularly in the islands of Sulawesi, Nusa Tenggara (including Bali), Maluku and Papua, and those who live across the Wallace line. The Wallace line runs through an area between the Sunda and Sahul plains, where the islands of Sulawesi, Nusa Tenggara and Maluku lie. This area is geographically unstable and is bounded by the deep sea with the two landmasses. Given its geographical location, this area serves as a faunal boundary of the two landmasses. Therefore, the Wallace line, plays a crucial role in the migration of Indonesian fauna between Sunda and Sahul landmasses. It also influenced the spread of humans. Indonesian territory is inhabited by two kinds of people namely the Australomelanesoid and the Mongoloid (Jacob 1967; 1974). The first live mostly in eastern Indonesia, the second reside in the western and northern parts of Indonesia. However, there is a mixture that blurs of the boundary between the two types, particularly in the area of the Wallace line, that serves as a central turning point between eastern and western Indonesia.

Research on human variation in the Southeast Asian islands (including the Wallacea area) has long been the subject of various strategies of scientific inquiry that urged researches in natural history, medicine, physical anthropology and genetics in the 19th and 20th centuries to explain variations in human types (Sysling 2019). The history of Western anthropologists research in the Wallace line region, namely in Nusa Tenggara (Timor, Flores and Sumba Islands) aimed to find out whether the Wallace line can more accurately define the measurements of human body. This research began in 1891 when the Dutch anthropologist Herman ten Kate conducted research on Flores and its surrounding islands (Semau, Roti, Sawu, Sumba, Solor and Adonara). Anthropometric measurements were conducted on 1,318 people at that time and revealed the characteristics of the Negro in the highlanders of Flores Island, despite the strong heterogeneity (the presence of other racial characteristics, such as Hindus and American Indians) and prominent differences with Sumbanese. After two decades, Hendrik Bijlmer, Doeke Brouwer and Wilhelmina Keers began studying the body size of humans living all over the Timor islands. Bijlmer measured hundreds of people in eight different places in Timor, Flores and Sumba and discovered Melanesian elements in the people of Timor and Flores, while

Melanesian influence was absent in Sumbanese, as was found earlier by ten Kate. In the 1930s, Doeke Brouwer, a military doctor residing on the island of Alor, east of Flores, studied the people of Alor and Pantar. Brouwer classified them in general as Melanesians with mostly Papuan characteristics and those with pre-Malay influences summarized (Sysling 2019). From these studies, Sysling (2019) concluded that there was still an unclear marker that can accurately measure the differences between Melanesian (Papuan) and Malay elements in the region. Hence, it is necessary to conduct research on the bodily characteristics of people in the Wallace Region to explain their variation.

This paper aims to present and discuss anthropometric characteristics of four indigenous populations or ethnic groups known as isolates living in Wallacea including the following: Naulu, Tana Ai, Bugis, and Rampasasa. Naulu live in the Rouhua hamlet, Sepa village, Masohi district, Central Maluku on the island of Seram, Maluku. Tana Ai live in the Tuabao hamlet area, Natarmage village, Talibura sub-district, Sikka Regency, East Nusa Tenggara. Bugis live in the hamlet of Salimbongan, Ulusaddang village, Lembang sub-district, Pinrang district, South Sulawesi, while Rampasasa people live in the Rampasasa hamlet, Waerii district, Waemulu villa, Manggarai regency on Flores Island.

Anthropometric characteristics are the easiest element to observe in individuals. Their variation is attributed to many factors including genetics and the environment, diet, physical activity, geography, lifestyle, disease and socioeconomic conditions (Lin et al. 2004; Yusof et al. 2007; Ashizawa et al. 2009; Iseri and Arslan 2009; Chuan et al. 2010; Karmegam et al. 2011; Luo et al. 2019; Funk et al. 2020). Several studies highlighted significant variations in several body dimensions in different nations, and ethnicities. Lin et al. (2004) compared 33 body dimensions and 31 body proportions in 4 different ethnicities in East Asia, namely the Chinese, Japanese, Korean and Taiwanese populations. The research shows that there are ethnic differences in the variation of body dimensions and proportions. Likewise, Khadem and Islam (2014) in their study of the male Bangladeshi population found significant differences in the body shape of male populations in different countries, such as India, Sri Lanka, Singapore, Portugal, and the Netherlands. However, Iseri and Arslan (2009) in their research on the Turkish population from 7 different geographical areas found that despite their relatively close ethnicity and substantially different cultures and social conditions, there were no considerable differences in the 36 anthropometric measurements taken, except for very significant weight differences. Meanwhile, in the Indonesian population, a study conducted by Wibowo et al. (2012) on Javanese and Madurese showed that the average body of Javanese and Madurese male farmers were smaller than those of Indian and Thai male farmers. Chuan et al. (2010) compared 37 body dimensions between Indonesians and Singaporeans and disclosed that there were significant differences between the 37 body dimensions in both populations.

Regional variations in anthropometric measurements are always present, even among people within an ethnic group. Widyanti et al. (2015) measured 38 body dimensions in 3 major ethnicities in Indonesia, namely the Minangkabau, Sudanese (West Java), and Javanese (Central Java and East Java) and identified significant differences in several body dimensions and proportions between these populations. This research supports the ethnic differences in anthropometric data in Indonesia. Currently, there are still few ethnic anthropometric databases in Indonesia because there are only few studies on anthropometry concerning ethnicity and indigenous people. This condition highlights the need to study the anthropometric characteristics of the four indigenous populations namely the Naulu, Tana Ai, Bugis and Rampasasa, who live in the Wallace line area, as a way to complement the need for ethnic anthropometric data in Indonesia. It is expected that this database can serve as a reference for ethnic anthropometric data in Indonesia in tracing the ancestors of eastern Indonesians, as well as comparison to ethnic groups in other countries.

Materials and methods

The research was conducted in four areas of East Indonesia in 2005 (Figure 1). The research subjects were 484 indigenous peoples consisting of 219 males and 265 females, aged between 18 and 80 years. They were divided into 4 samples:

1. The Naulu consisting of 62 males and 53 females.
2. The Tana Ai consisting of 54 males and 70 females.
3. The Bugis consisting of 65 males and 99 females.
4. The Rampasasa consisting of 38 males and 43 females.

The Naulu population lives in Amahai Regency in Seram Island, Central Maluku, the population of Tana Ai in Sikka, East Nusa Tenggara, the Bugis population in Pinrang Regency, South Sulawesi, and the Rampasasa population in Waerii Waemulu village, Manggarai Regency of West Flores.

Anthropometric measurements

Anthropometric measurements of all subjects were taken by including height (vertical distance from the vertex to the floor with the subject standing upright, using an anthropometer with an accuracy of 1 mm), body weight (body mass measured when the subject was standing using a body weight scale with an accuracy of 0.1 kg), sitting height (vertical distance from the vertex to the seat with the subject sitting using an anthropometer with an accuracy of 1 mm), biacromial width (horizontal distance between the right and left acromial with the subject standing upright using a spreading caliper with an accuracy of 1 mm) and bicristal width (horizontal distance between right and left iliocristale with the subject standing upright using a spreading caliper with an accuracy of 1 mm), trunk length (vertical distance from the suprasternale point to the symphision point with the subject standing upright, using an anthropometer with an accuracy of 1 mm), and leg length (vertical distance from trochanterion point to floor with the subject standing upright, using an anthropometer with an accuracy of 1 mm). Anthropometric measurements were conducted according to the measurement instructions of Norton and Olds (2004). The anthropometric indices were calculated, including the cormic index, trunk index, biacromial index, bicristal index and acromio-iliac index (Olivier 1969), and body mass index (BMI) according to the World Health Organization (WHO) classification (2007). The study has been approved by the Research Ethics Committee, Faculty of Medicine, Public Health and Nursing Universitas Gadjah Mada.

Table 1. Anthropometric characteristics of males in Wallacea, East Indonesia

Variables	Naulu (62)	Tana Ai (54)	Bugis (65)	Rampasasa (38)
	Means ± SD	Means ± SD	Means ± SD	Means ± SD
Age (years)[a*, b**, f**]	35.3 ± 13.2[h]	41.6 ± 12.6[g]	46.6 ± 13.2[h]	38.9 ± 14.8
Weight (kg)[a**, c**, d**, f**]	52.6 ± 5.3[h]	45.6 ± 6.1[h]	51.6 ± 6.8[h]	44.6 ± 4.^{9}h
Standing height (cm)[b**, c**, d*, e**, f**]	158.9 ± 4.9[h]	157.9 ± 6.8[h]	155.8 ± 6.6[h]	150.1 ± 3.^{3}h
Sitting height (cm)[a**, b*, c**, d**, f**]	81.2 ± 2.^{8}h	77.9 ± 3.1[h]	79.7 ± 3.8[h]	76.9 ± 3.4
Shoulder breadth (cm)[a**, c**, d**, e**, f**]	35.4 ± 1.9[h]	31.4 ± 1.^{9}h	36.2 ± 3.8[h]	33.6 ± 1.6[h]
Hip breadth (cm)[b**, d**]	23.9 ± 1.5	23.7 ± 1.8	24.8 ± 1.9[h]	24.5 ± 1.^{8}h
Trunk Length (cm)[d*]	50.9 ± 3.3[h]	51.2 ± 4.2[h]	49.8 ± 3.4[h]	50.7 ± 3.4[h]
Leg Length (cm) [b**, c**, d**, e**, f**]	85.5 ± 4.1[h]	85.4 ± 4.3[h]	83.4 ± 4.3[g]	78.1 ± 3.^{1}h
BMI (kg/m^2) [a**, c**, d**, e**, f**]	20.8 ± 1.7	18.2 ± 1.9	21.2 ± 1.9	19.6 ± 1.8
Cormic Index (%) [a**, d**, e**]	51.1 ± 1.4	49.4 ± 1.7[h]	51.2 ± 1.6[h]	50.9 ± 1.6[h]
Trunk Index (%) [c**, e**, f**]	32.0 ± 1.8[h]	32.4 ± 2.0[h]	32.0 ± 1.7[h]	33.6 ± 1.9[h]
Acromial Index (%) [a**, b**, d**, e**, f**]	22.3 ± 1.0[h]	19.9 ± 1.2[h]	23.3 ± 1.5	22.2 ± 1.0[h]
Iliocristal Index (%) [b**, c**, d**, e**]	15.0 ± 0.9[h]	15.0 ± 1.1[g]	15.9 ± 1.2[h]	16.2 ± 1.1[h]
Acromiocristale Index (%) [a**, c**, d**, e**, f**]	67.7 ± 5.5[h]	75.6 ± 5.6[h]	68.9 ± 5.2	73.2 ± 6.7[h]

*$p<0.05$; **$p<0.01$; [a]difference between Naulu and Tana Ai; [b]difference between Naulu and Bugis; [c]difference between Naulu and Rampasasa; [d]difference between Tana Ai and Bugis; [e]difference between Tana Ai and Rampasasa; [f]difference between Bugis and Rampasa; BMI: Body mass index; [g]$p<0.05$ difference between male and female; [h]$p<0.01$ difference between males and females

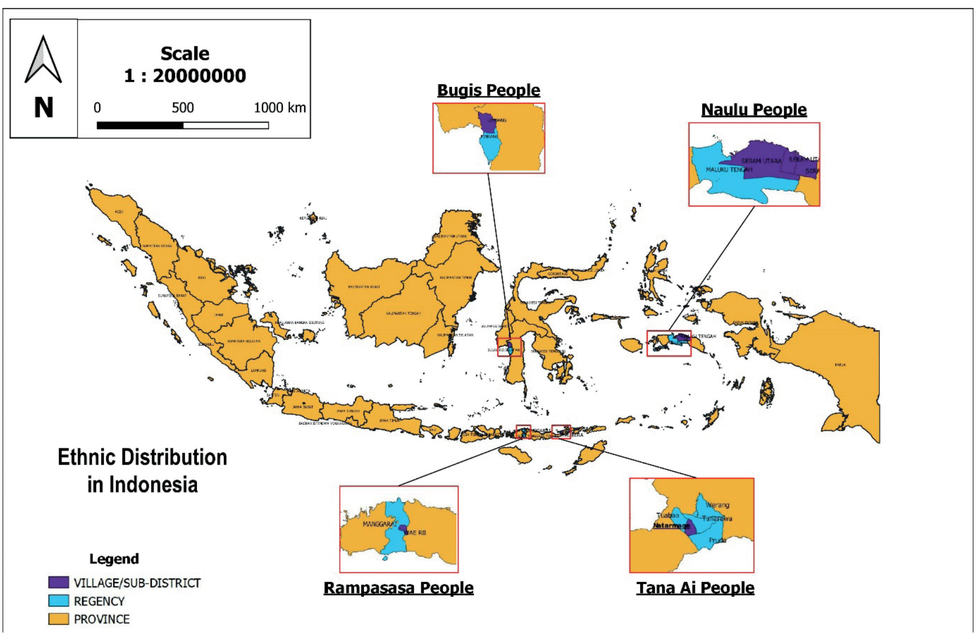

Fig. 1. Map of Indonesia

The one-way ANOVA was used for the statistical analysis with the LSD post hoc test to compare anthropometric characteristics between populations and the independent T-test to determine differences between the sexes in the population, with a significance value of $p < 0.05$.

Results

Descriptive statistical anthropometric data for the Naulu, Tana Ai, Bugis and Rampasasa tribes are listed in Tables 1 and 2, while the anthropometric indices data are shown in Tables 3 and 4. Of the 4 indigenous peoples, it was observed that the Tana Ai and Bugis males weighed significantly more ($p<0.001$) than the Naulu and Rampasasa males. The Naulu males had the average height (158.9 cm), while the Rampasasa males had the lowest measurements in terms of weight and height (44.6 kg and 150.1 cm), sitting height (76.9 cm), and leg length (78.1 cm). The smallest shoulder size (31.4 cm) was found in the Tana Ai males, while Naulu males had the largest sitting height measurement (81.2 cm). The male BMI category in the 4 populations showed normal nutritional status ranging from 18.2 to 21.2. At this point, it can be concluded that Naulu males have the largest body size, while Rampasasa males are the smallest (Table 1).

Table 2. Anthropometry characteristics of females in Wallace, East Indonesia

Variables	Naulu (53)	Tana Ai (70)	Bugis (99)	Rampasasa (43)
	Means ± SD	Means ± SD	Means ± SD	Means ± SD
Age (years)$^{a*, b**, c**}$	26.3 ± 6.3	35.8 ± 14.0	38.2 ± 13.2	40.0 ± 19.0
Weight (kg)$^{a**, c**, d**, f**}$	47.3 ± 6.9	39.6 ± 6.4	48.4 ± 7.7	40.4 ± 6.5
Standing height (cm)$^{c**, e**, f**}$	148.5 ± 3.9	148.3 ± 4.5	148.5 ± 5.2	143.4 ± 5.4
Sitting height (cm)$^{a**, b*, d**, f**}$	76.5 ± 2.7	74.9 ± 2.4	77.3 ± 3.1	75.8 ± 4.2
Shoulder breadth (cm)$^{a**, c**, d**, e**, f*}$	31.4 ± 1.6	27.9 ± 1.9	30.9 ± 2.0	30.2 ± 1.7
Hip breadth (cm) $^{a*, b*, d**, e**, f**}$	23.8 ± 1.8	23.0 ± 2.0	23.1 ± 2.1	25.9 ± 1.8
Trunk Length (cm) $^{a**, b**, c**, e**, f**}$	41.8 ± 2.1	45.4 ± 2.9	47.8 ± 3.3	44.6 ± 3.6
Leg Length (cm) $^{a*, c**, e**, f**}$	82.9 ± 2.8	81.4 ± 3.6	82.1 ± 3.3	75.7 ± 3.3
BMI (kg/m^2) $^{a**, c**, d**, e**, f**}$	21.4 ± 3.0	17.9 ± 2.5	21.9 ± 3.2	19.6 ± 2.6
Cormic Index (%) $^{a**, c**, d**, e**, f**}$	51.5 ± 1.4	50.6 ± 1.5	52.0 ± 1.3	52.8 ± 1.8
Trunk Index (%) $^{a**, b**, c**, e**, f**}$	28.2 ± 1.3	30.6 ± 1.7	32.2 ± 1.8	31.1 ± 1.9
Acromial Index (%) $^{a**, d**, e**}$	21.1 ± 1.0	18.9 ± 1.1	20.8 ± 1.2	21.1 ± 1.1
Iliocristal Index (%) $^{a*, b*, c**}$	16.1 ± 1.2	15.5 ± 1.3	15.6 ± 1.4	18.1 ± 1.2
Acromiocristale Index (%) $^{a**, c**, d**, e**, f**}$	76.1 ± 4.9	82.6 ± 7.7	74.8 ± 6.7	86.0 ± 6.4

*p<0.05; **p<0.01; [a]difference between Naulu and Tana Ai; [b]difference between Naulu and Bugis; [c]difference between Naulu and Rampasasa; [d]difference between Tana Ai and Bugis; [e]difference between Tana Ai and Rampasasa; [f]difference between Bugis and Rampasa; BMI: Body mass index; [g]p<0.05 difference between male and female; [h]p<0.01 difference between males and females

The anthropometric data for the female group in these four populations indicate that the Rampasasa females had the shortest height (143.4 cm) and leg length (75.7 cm), but had the widest hip size (25.9 cm). The smallest shoulder size was found in the Tana Ai female (27.9 cm), the shortest trunk was in the Naulu female (41.8 cm), while the longest trunk was in the Bugis female (47.8 cm). The BMI category in the females of the four populations showed normal nutritional status, except for the Tana Ai females who were classified in the underweight category (Table 2). At this point, it can be said that the Rampasasa females had the smallest body size, while the Tana Ai females had a poor nutritional status (Table 2).

Sexual dimorphism was evident in every population in this study as seen from the significant differences between males and females from the 4 populations. Significant differences ($p<0.01$) between males and females in the same population were found in height, weight, sitting height (except for the Rampasasa tribe), shoulder width, hip width (except for the Tana Ai and Naulu tribes), trunk length, leg length, cormic index (except for the Naulu tribe), trunk index (except for the Bugis tribe), acromial index, iliocristalis index (except for the Bugis tribe), and acromiocristalis index ($p<0.001$).

Table 3. Anthropometric indices categories of males in Wallace, East Indonesia

Variables	Naulu (62) N (%)	Tana Ai (54) N (%)	Bugis (65) N (%)	Rampasasa (38) N (%)
BMI (kg/m^2)				
Underweight	4 (6.5%)	22 (40.7%)	3 (4.6%)	8 (21.1%)
Normal	50 (80.6%)	31 (57.4%)	50 (76.9%)	27 (71.1%)
Obesity	8 (12.9%)	1 (1.9%)	12 (18.5%)	3 (7.8%)
Cormic Index (%)				
Brachycorm	26 (41.9%)	44 (81.4%)	30 (46.2%)	18 (47.4%)
Mesocorm	33 (53.2%)	9 (16.7%)	24 (36.9%)	17 (44.7%)
Macrocorm	3 (4.9%)	1 (1.9%)	11 (16.9%)	3 (7.9%)
Trunk Index (%)				
Short trunk	1 (1.6%)	0	0	0
Medium trunk	5 (8.1%)	6 (11.1%)	7 (10.8%)	2 (5.3%)
Long trunk	56 (90.3%)	48 (88.9%)	58 (89.2%)	36 (94.7%)
Acromial Index (%)				
Narrow	22 (35.5%)	52 (96.3%)	7 (10.7%)	12 (31.6%)
Medium	25 (40.3%)	2 (3.7%)	20 (30.8%)	17 (44.7%)
Broad	15 (24.2%)	0	38 (58.5%)	9 (23.7%)
Iliocristal Index (%)				
Dolichopelvic	54 (87.1%)	43 (79.6%)	32 (49.2%)	14 (36.8%)
Mesopelvic	8 (12.9%)	10 (18.5%)	26 (40.0%)	23 (60.6%)
Brachypelvic	0	1 (1.9%)	7 (10.8%)	1 (2.6%)
Acromiocristale Index (%)				
Trapezoidal trunk	42(67.7%)	8 (14.8%)	43 (66.2%)	6 (15.8%)
Intermediate trunk	15 (24.2%)	14 (25.9%)	11 (16.9%)	15 (39.5%)
Rectangular trunk	5 (8.1%)	32 (59.2%)	11 (16.9%)	17 (44.7%)

Table 4. Anthropometric indices categories of females in Wallace, East Indonesia

Variables	Naulu (53) N (%)	Tana Ai (70) N (%)	Bugis (99) N (%)	Rampasasa (43) N (%)
BMI (kg/m^2)				
Underweight	5 (9.4%0	36 (51.4%)	6 (6.1%)	12 (27.9%)
Normal	33 (62.3%)	32 (45.7%)	63 (63.6%)	27 (62.8%)
Obesity	15 (28.3%)	2 (2.9%)	30 (30.3%)	4 (9.3%)
Cormic Index (%)				
Brachycorm	21 (39.6%)	48 (68.6%)	27 (27.3%)	11 (25.6%)
Mesocorm	13(24.5%)	10 (14.3%)	24 (24.2%)	2 (4.6%)
Macrocorm	19 (35.8%)	12 (17.1%)	48 (48.5%)	30 (69.8%)
Trunk Index (%)				
Short trunk	24 (45.3%)	1 (1.4%)	2 (2.0%)	2 (4.7%)
Medium trunk	26 (49.1%)	24 (34.3%)	5 (5.1%)	8 (18.6%)
Long trunk	3 (5.6%)	45 (64.3%)	92 (92.9%)	33 (76.7%)
Acromial Index (%)				
Narrow	44 (83.0%)	70 (100%)	81 (81.8%)	33 (76.8%)
Medium	8 (15.1%)	0	14 (14.1%)	9 (20.9%)
Broad	1 (1.9%)	0	4 (4.1%)	1 (2.3%)
Iliocristal Index (%)				
Dolichopelvic	19 (35.8%)	43 (61.4%)	62 (62.6%)	0
Mesopelvic	34 (64.2%)	23 (32.9%)	33 (33.3%)	18 (41.9%)
Brachypelvic	0	4 (5.7%)	4 (4.1%)	25 (58.1%)
Acromiocristale Index (%)				
Trapezoidal trunk	5 (9.4%)	3 (4.3%)	22 (22.2%)	0
Intermediate trunk	15 (28.3%)	8 (11.4%)	30 (30.3%)	1 (2.3%)
Rectangular trunk	33 (62.3%)	59 (84.2%)	47 (47.5%)	42 (97.7%)

The anthropometric indices data show that Tana Ai males had the largest percentage of underweight (40.7%) compared to the other 3 tribes, while the highest obesity rate was found in Bugis males (18.5%). The cormic index as the ratio between sitting height and body height, indicated that Tana Ai males had the highest percentage of the brachycorm category (short trunk and long limbs) (81.4%), and the mesocorm category (16.7%) and the least macrocorm (long trunk size, short limbs) (1.9%) compared to the other 3 tribes. The biacromial index, which describes the width of the shoulders, showed that narrow shoulders with the highest percentage were found in Tana Ai males (96.3%), while the wide shoulders were mostly found in Bugis males (58.5%). The iliocristale index, which describes hip width, highlighted that on average men from the 4 populations had a narrow hip category, except for Rampasasa males who had the highest category of medium hips (60.6%). In terms of the shape of the trunk, it can be seen that there was a significant variation in the shape of the trunk between the 4 populations with a similar pattern between Naulu and Bugis males, namely the trapezoidal type, while the trunk shape of the Tana Ai and Rampasasa males was rectangular (Table 3).

Similar to Tana Ai males, it was proven that Tana Ai females had the largest percentage of underweight (51.4%) compared to the other 3 tribes, while the highest obesity rate was found in Bugis females (30.3%), which was also true for Bugis males. The cormic index showed that Tana Ai females had the highest percentage of brachycorm (68.6%), while the highest percentage of macrocorm was found in Rampasasa females (69.8%). Bugis females had the longest trunks (92.9%), while Naulu females had the shortest trunks (45.3%). In the acromial index, most females from the 3 populations had narrow shoulders, except for the Tana Ai tribe, who had narrow shoulders (100%). In terms of hip size or bicristal index, Rampasasa females had the widest hips (58.1%) compared to the other 3 populations. As for the average trunk shape, most of the women in these 4 populations were of the rectangular type (Table 4).

Discussion

Based on the statistical analysis, generally, there were different anthropometric sizes in the four indigenous peoples living in Eastern Indonesia. These differences include height and weight, shoulders and hips, trunk and legs as well as anthropometric indices. In terms of height, the most striking difference was in the height of the Rampasasa people, who were clearly shorter than the other 3 populations. This is understandable because the Rampasasa people in this study belonged to the Pygmy group (Jacob et al. 2008) with an average male height of 150.1 cm, while Rampasasa female had 143.4 cm. Generally, the Pygmy who live in Africa, the Philippines, Malaysia and Papua New Guinea have an average adult height of 150–155 cm, (Ishida et al. 1998; Jacob et al. 2006; Funk et al. 2020). According to Froment (2014) the average height of the Pygmy group varied from 142 cm (Western Congo Basin Efe) to 161 cm (Eastern Twa). Meanwhile, the size of the shoulders and pelvis of the Rampasasa males as compared to the Pygmy males of Western New Guinea (Ponzetta et al. 2013) was smaller.

The Naulu and Tana Ai males had almost the same height and were larger than the Bugis and Rampasasa males. The result of NCD Risk Factor Collaboration study (2016) demonstrated that Indonesian male adults had an average height of around 160 cm. This figure is categorized as short average height. Of the 200 countries whose average height was measured, Indonesians were ranked 188th, below Papua New Guinea, Myanmar and Vietnam. The height of Indonesian people was varied, between 135 and 180 cm. In other words, it ranges from short to tall. The average height may vary slightly between some local populations, but in most cases the differences are insignificant, and the variations in the current Indonesian population can be studied more deeply, because the variations are indeed deemed extraordinary, and are influenced by monogenic and polygenic characteristics, morphological, physiological, serological and biochemical characteristics (Jacob 1974).

Human populations all around the world show marked variations in body size (Little et al. 2020). The differences between the 4 populations can be clearly seen as a description of different geographical conditions. Even though they all live in the Wallace line area, because they are located in different geographical areas, they have different body characteristics. This situation is supported by Sirajuddin et al. (1994) in their research, which stated that geographical factors had a genetic influence on the distinctive features of ethnic groups living in southern India.

The Naulu people mostly live on the coast with farming and fishing as main occupations. Their staple foods are especially cassava, sweet potatoes, bananas and sago, which they consume daily. Meanwhile, the Tana Ai people live in the village, which is known as a forest and hilly area. Apart from farming, they also do gardening as the main source of livelihood. Their main sources of food are rice, corn, peanuts and cassava. The Bugis people live in the hills in the villages. Their main source of livelihood is farming and livestock husbandry (Hidayah 1996). The Rampasasa people live in the hills, their main occupation is farming using crop cultivation systems, such as corn, cassava, sweet potatoes, and red beans. These people eat meat only during traditional ceremonies, festivals and weddings and also during times of mourning (Jacob et al. 2008). In terms of the body dimensions based on sex, it is apparent that the Naulu male had the largest average size of body weight, height, sitting height and leg length compared to the 3 other groups of male tribes. This situation is understandable considering that the Naulu people live on the coast, allowing them to have adequate protein and more abundant food sources than the other 3 populations who all live in hilly and forest areas (Hidayah 1996; Jacob et al. 2008).

The results of this study are in line with several studies, including research by Iseri and Arslan (2009) on the Turkish population, which disclosed that geographical conditions are characterized by ethnic uniqueness with different habits, nutritional intake, and socio-economic conditions. All of these factors significantly affect height and weight. Apart from geographical influences, dietary factors and secular changes also affect anthropometric data, which is in accordance with the statement of Chuan et al. (2010) that nutritional status affected the growth of adult bodies in the population. These findings also parallel the results of research by Ashizawa et al. (2009) showing that there were secular changes that affected the anthropometric differences between the Javanese and Chinese populations.

Naulu females had the widest shoulders and longest legs, while Bugis females had the highest average body weight, sitting height and trunk length compared to females from the other 3 ethnic groups. On the other hand, the Tana Ai females, aside from having the smallest weight and sitting height, also had the narrowest shoulders among the females of the other 3 tribes. When compared with research by Lin et al. (2004), the height, sitting height, shoulder width and hip width of the Naulu, Tana Ai, Bugis and Rampasa people were smaller than those of Taiwan, China, Japan and Korea. There are significant differences as shown by the comparison of the average height of Naulu male (158.9 cm) and the lowest average height of Chinese male (167.8 cm) in Lin et al.'s study (2004). In contrast, the Naulu male had the tallest average height among the 4 ethnic groups in this study. This is also supported by the research results of Glinka et al. (2010) where the anthropometric data of the Palue people (Australomelanesoid) were significantly smaller ($p<0.001$) compared to the Javanese (Mongoloid). Likewise, when compared with 3 other ethnic groups in Indonesia (Minangkabau, Sundanese and Javanese) who live in the western part of Indonesia (Widyanti et al. 2015), it was clear that on average, both the males and females of the Naulu, Tana Ai, Bugis and Rampasasa people, had smaller body size. This fact proves that genetic factors, economic development, social environment, and type of work affect body size in different ethnicities (Lin et al. 2004; Sysling 2019). In addition, the differences are also attributed to the fact that the 4 populations (Naulu, Tana Ai, Bugis and Rampasasa) live in isolation and inhabit ecosystems with limited food sources, which explains why they have a smaller average body size.

Comparison of anthropometric indices indicates that the 4 populations had long trunks, narrow shoulders and pelvis except for the Bugis, with a trapezoidal trunk except for the Tana Ai and Rampasasa people who mostly had a rectangular type. According to Olivier (1969), the trapezoidal type trunk is usually found in males, while the rectangular type is in females. A person who has a greater height tends to have a smaller index and is therefore more masculine.

In terms of sex differences in the same population, in general, this study showed that the anthropometric dimensions of males are larger and significantly different from females, except for the width of the hips. These results are supported by research by Widyanti et al. (2015) and Chuan et al. (2010), which showed that the dimensions of the male body are larger than the female. In line with this, Gasser et al. (2000), also held that it is commonly known that on average, adult males have a larger physical size than that of the females except for hip width. This is evidenced by studies of sexual dimorphism in height, sitting height, biacromial and bicristal dimensions.

This study contains limitations that are important to acknowledge. In the data collection, only occupation and medical history were used when collecting information from subjects. It is important to collect data on subjects' nutrition, genetics, physical activity, and culture at a wider scope. Further research is needed to address the aspect of different body sizes considering developments in the field of technology, transportation and large-scale migration, which may affect the demographic and physical characteristics of ethnic groups in Indonesia.

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Conflict of interest

The authors declare that there is no conflict of interest regarding the publication.

Authors' contributions

NTR was the lead researcher, conceived concept and design, article writing, critical revision of the article for important intellectual content, provision of study materials, data collection; JH performed the statistical analysis, data collection, and wrote the manuscript; RAS performed data collection and compilation, critical revision of the article for important intellectual content. All authors discussed the results and contributed to the final manuscript for publication.

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Birth Size of Neonates and Its Association with Seasonality

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Abstract: The aim of the study is to evaluate the relationship between the season of birth and the birth weight and length of Bulgarian newborns.

The weight and length data of 6517 (6098 full-term and 419 preterm) live births in 2000–2001 were collected from the birth registry of II Hospital of Obstetrics and Gynaecology "Sheynovo", Sofia, Bulgaria. Statistical analyses were done using the SPSS 16 software for Windows: descriptive statistics; the t-test ($p<0.05$); One-Way ANOVA, (Tukey, HSD-test, $p<0.05$) and the Pearson's correlation. The sunshine duration data for 2000 and 2001 were collected at the Sofia Meteorological Station.

The mean weight of Bulgarian neonates born in 2000-2001 was 3389.8 g in boys and 3261.8 g in girls. The average newborn's length was 51.0 cm and 50.3 cm in boys and girls, respectively. In all seasons, significant gender differences were observed with a priority for boys ($p\leq0.001$). The winter period was identified with a peak in birth length for both sexes, and spring and summer were the seasons with the lowest values for boys and girls, respectively. A significant positive correlation between birth length and the daily amount of sunshine during the prenatal period was found ($p<0.001$).

Seasonal fluctuations influenced weight and length in Bulgarian neonates. The results obtained in this study can be useful in prenatal diagnostics, neonatal care, and health prevention of pregnant women and neonates.

Key words: birth weight, birth length, seasonality.

Original article

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Introduction

Birth weight and length are the most important determinants of infant health status. Fetal development and newborn anthropometric dimensions depend on a mix of genetic, socio-economic and environmental factors, including the meteorological parameters of the surrounding environment (e.g., solar radiation, air pressure, temperature and humidity, wind speed, air pollution) mediated by the mothers (Wells and Cole 2002; Vershubskaya et al. 2016). In last centuries, the climate is constantly changing (Global Climate Change, NASA). This was especially true in the last decades of the 20th century and continues today, when climate change follows steady one-way trends rather than fluctuations around a certain average (Ways the Climate Changed Over the Past Decade). The upward trend in global temperature from 1880 to the present is 0.08°C per decade. However, in the last 40 years, the average global temperature has risen twice as fast and has a trend of 0.18°C per decade (Annual 2021 Global Climate Report, NOAA). Locally, global climate change is often undergoing transformation. This is due to the influence of local natural and anthropogenic factors, which determine specific features in the nature of climate change. This is especially true in urban areas, where a large part of the population is concentrated. In urban areas climate change is becoming even more noticeable as a result of the highly transformed surface and considerable air pollution. Currently registered climate change has a significant impact on people's working, living and recovery conditions. This affects all areas of human activity – agriculture, biodiversity and the forestry sector, water, energy, transport, tourism, as well as human health (Climate risk and response: Physical hazards and socioeconomic impacts, Report, January 2020). The question is no longer whether the change of climate exists, but to what extent it affects nature, man, human activity and health as well as how to mitigate and adapt to it.

Climate change has both direct and indirect impact on prenatal, neonatal, and postneonatal infant health (The Impact of Climate Change on Maternal and Newborn Health Outcomes). For temperate-continental latitudes, the meteorological elements have a well-expressed intra-annual course, which has a corresponding impact on the sizes of the newborns, as well as on some negative birth outcomes, such as low birth weight, malnutrition, and respiratory diseases (McGrath et al. 2005; Chersich et al. 2020). Sunshine duration, as an indicator of solar radiation, is one of the most important climate factors influencing the bio status of man. It affects the human body in two main ways: as a factor in heat exchange between the human body and the environment, and as a source of sunlight, including of ultraviolet (UV) radiation. UV rays are the most biologically active part of the light spectrum affecting blood and lymph circulation, erythropoiesis, neuro-reflex mechanisms, calcium-phosphorus exchange, metabolism, the formation of vitamin D (DeLuca 1986; Kumar 1986; Ross et al. 2011). Seasonal changes in maternal circulating 25-hydroxyvitamin D levels (25(OH)D), directly influence the levels of fetal vitamin D. Neonates born during the summer have almost two-fold higher levels of circulating 25-hydroxyvitamin D3 compared to those born in the winter (McGrath et al. 2005). Therefore, sunshine is a vital climate factor affecting health, especially in newborns, where mother's exposure conditions during pregnancy and the first stages of direct contact of the newborn with the environment are important.

We suppose that sunlight is one of the climate factors that influence prenatal development, which is manifested in the variability in birth sizes related to the birth season. The aim of the present study is to evaluate the relationship between the season of birth and the birth weight and length of Bulgarian newborns. For this purpose, we tracked the intra-annual course of the size of newborns in the capital city of Bulgaria – Sofia, as well as the intra-annual course of sunshine, as one of the most significant factors in bio-climatic terms.

Materials and Methods

The data of 6517 (6098 (93.6%) full-term and 419 (6.4%) preterm) live births in 2000–2001 were collected from the birth registry of II Hospital of Obstetrics and Gynaecology "Sheynovo", Sofia, Bulgaria. In the study, only data from full-term newborns (body weight of more than 2500 g) were analyzed. The babies were distributed according to the seasons of birth, defined as follows: winter (January-February-March), spring (April-May-June), summer (July-August-September), and autumn (October-November-December). The raw data sources we use in this study are organized according to a calendar pattern of the seasons, taking into account the influence of the specific latitude and the specific microclimate of Sofia. Climatic changes, which are well expressed at the geographical latitudes of Sofia, cause a certain transformation of the seasons, reflecting their onset, duration and typical characteristics. In recent decades, we have witnessed a very diverse manifestation of the seasons, dotted with significant deviations from the norm, which, to a certain extent, veiled their characteristic, astronomically determined features. The climate of Sofia is particularly strongly influenced by a number of local factors, such as morphography, orography and underlying surface. The basin-like character of the relief, the orographic closure of the Horizon from the south, as well as the highly anthropogenic underlying surface, generate highly expressed local modifications of the climate, reflecting, to some extent, the astronomical predetermination of the local climate in this area.

The conception date and accordingly the three trimesters of pregnancy were determined by the date of birth.

Statistical analysis of anthropometric data was performed using the Statistical package for social science (SPSS 16). The descriptive statistics, the t-test ($p<0.05$) and One-Way ANOVA, as well as the post hoc procedures for multiple comparisons (Tukey HSD test; $p<0.05$) were implemented. The Pearson correlation analysis ($p<0.05/0.001$) was used to assess the relationship between the anthropometric features of neonates and sunshine duration in the three trimesters of pregnancy by the season of birth. Children born before October 2000 were excluded from the analysis because we do not have data about the sunshine duration for 1999.

Data on the duration of sunshine in the years 2000 and 2001 were collected at the Sofia Meteorological Station. The data were generated through direct in-situ ground observations, with standard, calibrated instruments, in the system of the National Meteorological Network, and were obtained from the meteorological archive of the National Institute of Meteorology and Hydrology (NIMH). For the purposes of the present study, a database of daily sums of sunshine duration (in hours) was created for the period 2000–2001 (corresponding to the data on newborns in the capital city in 2000 and 2001, the same period).

Results

The average annual duration of sunshine in Sofia is about 2200 hours (Mateeva 1999). This is slightly over 50% of its astronomically possible average annual duration (Fig. 1). Compared to most stations from the southern part of the country and many other stations located in Northern Bulgaria, Sofia has a shorter duration of sunshine (Mateeva 1989). The reason for this might be a significant orographic shadowing of the horizon by the mountains located to the south, southeast and southwest observed in the capital city of Sofia. In addition, its low-lying position, at the bottom of Sofia hollow, causes frequent formation of inversion conditions of the atmosphere during the cold half-year and, accordingly, an increased number of days with fog (Mateeva 2002).

The years 2000 and 2001 were distinguished by higher than average multi-annual values of sunshine. This was especially true for the year 2000, for which the annual sum of sunshine hours reached 2611 h. This is due to a very sunny spring, summer and December-January period in that year. For comparison, in 2001, the annual amount of sunshine hours were normal (2246 h).

The monthly sums of sunshine in the two observed years were characterized by a well-expressed seasonal course with a maximum in July and a minimum in

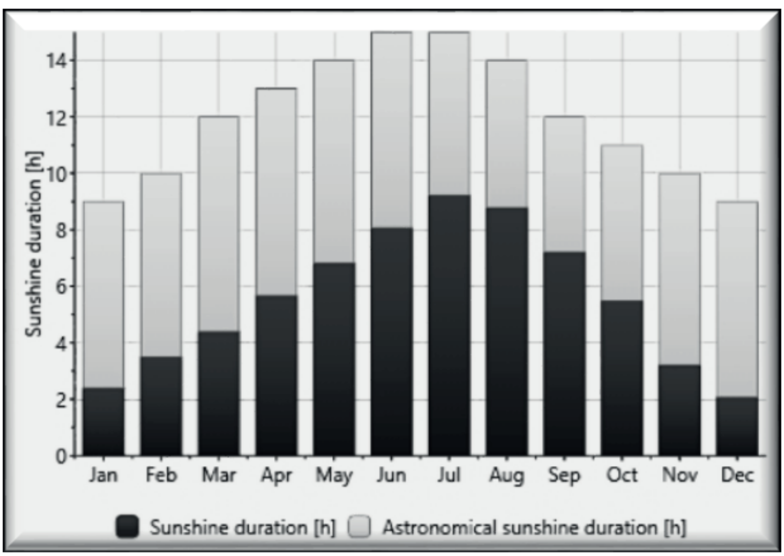

Fig. 1. Intra-annual course of the average daily duration of sunshine compared to its astronomically possible duration in Sofia (Source: Worldwide Irradiation Data). Available online: https://meteonorm.com/en/

December. The summer amount of sunshine hours represented about 39–40% of the annual amount, while in winter it was only 10–12%. During the transition seasons, the share of sunny hours was about 25%. In the year 2000, a certain anomaly is observed – the autumn amount of sunshine hours was less than the spring, and the winter and summer amounts were unusually high (Fig. 2).

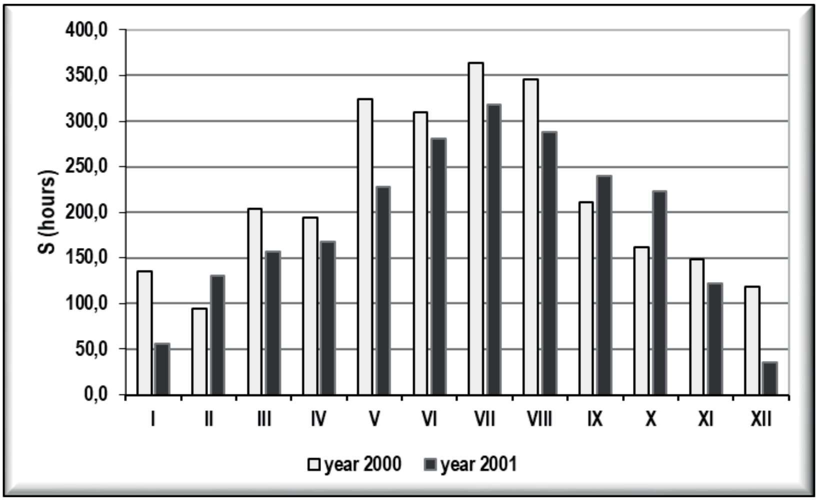

Fig. 2. Intra-annual course of the monthly duration of sunshine (S) in Sofia for 2000 and 2001

Table 1 shows the statistical data in full-term newborns by the season of birth. The number of live births varied considerably by the season, with most births occurring in the summer months, and the lowest in winter (for boys) and autumn (for girls). In all seasons, the number of male newborns exceeded the number of females. The mean values of weight in neonates born in 2000–2001 were 3389.8 g in boys and 3261.8 g in girls. The average newborn's length was 51.0 cm and 50.3 cm in boys and girls, respectively. In all seasons, significant sexual differences according to the newborn sizes with priority for boys were observed ($p \leq 0.001$). Specifically, they were heavier and longer than girls with 128.1 g and 0.74 cm, respectively. Analysis of variance detected significant influences of the season of birth on the length of boys ($F=7.49$; $p<0.001$) and girls ($F=2.78$; $p<0.05$). The winter period was identified with a peak in birth length in both sexes (Figure 3). The lowest values of birth length were established in spring for boys and in summer for girls. Seasonal variation in the boys' length was expressed with appreciably higher mean values in winter than in spring ($p<0.001$). The girls born in winter were significantly longer than those born in summer and autumn months ($p \leq 0.001$). Annual variability in birth weight in both sexes was also found, as autumn-born boys, spring-born girls were heavier, and the winter-born boys and summer-born girls were lighter. However, this effect was statistically insignificant.

(A)

(B)

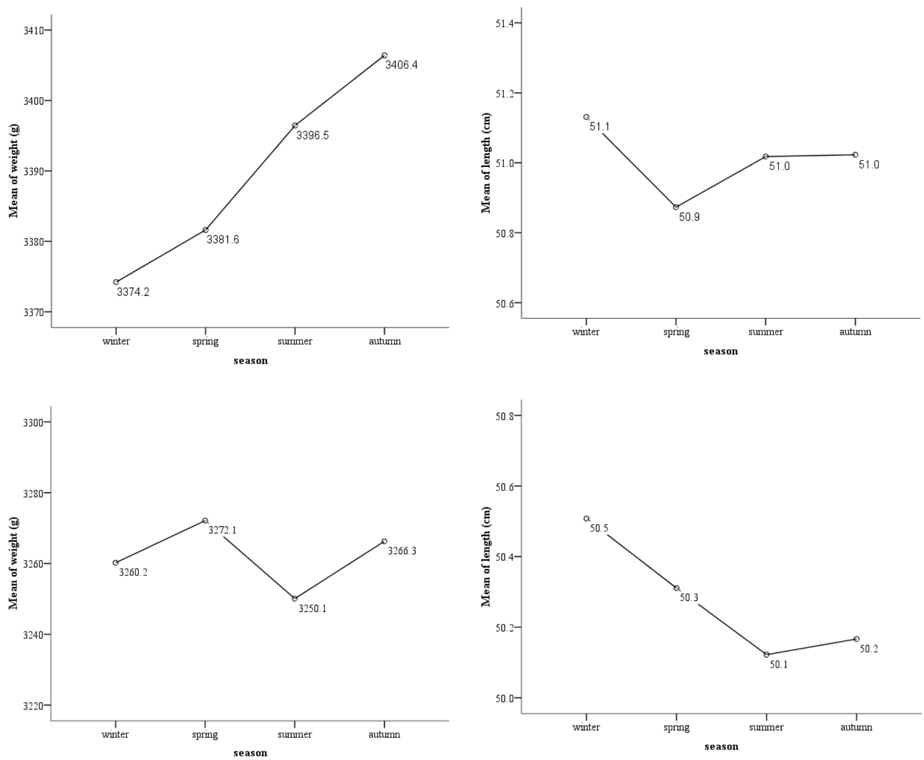

Fig. 3. Average birth weight and length by season in newborn boys (A) and girls (B)

Table 1. The mean values of birth weight and length in full-term neonates, according to the season of birth

Features	Birth year	2000-2001						p-value (♂/♀)
	Season of birth	boys			girls			
		n	mean	SD	n	mean	SD	
Birth weight	winter	774	3374.22	418.99	705	3260.20	390.58	0.001
	spring	780	3381.62	408.51	724	3272.11	384.82	0.001
	summer	838	3396.46	420.16	810	3250.07	382.91	0.001
	autumn	783	3406.41	424.03	684	3266.27	398.74	0.001
	Total	3175	3389.85	417.99	2923	3261.77	388.87	0.001
Birth length	winter	771	51.13[b]	1.84	705	50.51[c, d]	1.72	0.001
	spring	780	50.87[a]	1.72	724	50.31	1.67	0.001
	summer	838	51.02	1.74	810	50.12[a]	1.78	0.001
	autumn	783	51.02	1.77	684	50.17[a]	1.68	0.001
	Total	3172	51.01	1.77	2923	50.27	1.72	0.001

ANOVA Tukey HSD test: **a**-significant differences compared with winter newborns; **b**- significant differences compared with spring newborns; **c**- significant differences compared with summer newborns; d- significant differences compared with autumn newborns;

Table 2 presents the strength of the relationship between the average sum of sunlight during the three trimesters of prenatal growth and the birth sizes, according to the season of birth. The length of newborn boys correlated significantly with the daily sum of sunshine in the second trimester of pregnancy although this relationship was weaker ($r=0.109$, $p<0.001$). Weak positive ($r=0.120$) and statistically significant ($p=0.019$) correlation was found between the birth weight of boys born in the winter and the sunshine duration in the second trimester of pregnancy. In newborn girls, positive weak correlation between the sunshine duration in the first ($r=0.130$, $p<0.001$) and second ($r=0.105$, $p<0.001$) trimesters of pregnancy and birth' length was also found, especially in those born in spring ($p=0.033$; $p=0.018$) and autumn ($p=0.003$; $p<0.001$). Weak positive correlation was also observed between the birth weight of girls and sunlight exposure during the first trimester of pregnancy ($r=0.109$, $p=0.044$) but only in those born in winter. In addition, in newborn girls, statistically significant negative correlation was observed between their height and weight and sun exposure of the mother during the second and third trimesters of pregnancy.

Table 2. Correlation between newborn birth sizes and sunshine duration across the three trimesters of prenatal growth by seasons of birth.

Trimesters Sunshine duration	Boys (n=1946)					Girls (n=1819)				
		Birth weight		Birth length			Birth weight		Birth length	
	n	r	p	r	p	n	r	p	r	p
Winter										
1st	383	-0.075	0.141	-0.025	0.628	345	**0.109***	**0.044**	-0.017	0.758
2nd	383	**0.120***	**0.019**	0.093	0.070	345	**-0.118***	**0.028**	0.021	0.695
3rd	383	0.072	0.160	0.020	0.695	345	**-0.107***	**0.046**	0.016	0.764
Spring										
1st	394	0.002	0.963	-0.004	0.943	386	0.075	0.140	**0.108***	**0.033**
2nd	394	0.001	0.980	-0.016	0.756	386	0.094	0.064	**0.121***	**0.018**
3rd	394	-0.002	0.965	0.005	0.918	386	-0.078	0.125	**-0.111***	**0.029**
Summer										
1st	386	-0.041	0.427	-0.062	0.228	404	0.062	0.216	0.061	0.224
2nd	386	0.039	0.440	0.060	0.239	404	-0.061	0.222	-0.046	0.356
3rd	386	0.041	0.425	0.062	0.225	404	-0.062	0.211	-0.055	0.273
Autumn										
1st	783	-0.036	0.316	0.002	0.965	684	0.043	0.266	**0.114****	0.003
2nd	783	-0.050	0.159	0.027	0.450	684	0.062	0.105	**0.143****	**0.001**
3rd	783	0.020	0.577	0.028	0.430	684	-0.014	0.710	-0.055	0.150
Total /October 2000-December 2001/										
1st	1946	-0.039	0.084	0.011	0.632	1819	0.044	0.062	**0.130****	**0.001**
2nd	1946	0.017	0.461	**0.109****	**0.001**	1819	0.006	0.796	**0.105****	**0.001**
3rd	1946	0.037	0.103	0.019	0.405	1819	-0.035	0.140	**-0.098****	**0.001**

*. Statistically significance at $p \leq 0.05$; **. Statistically significance at $p \leq 0.01$;

Discussion

The present study focuses on the seasonality in weight and length of neonates born at the beginning of the XXIth century in Sofia, as well as on the influence of the sunshine duration during pregnancy on both characteristics.

The meteorological elements and socio-demographic factors have a corresponding impact on the reproductive function of the human and, in particular, on the seasonal variation of birth rates (Kumari and Venkateswar 1982; Bobak and Gjonca 2001). The seasonal tendency of births during the investigated years (2000–2001), which is characterized by a peak in summer and troughs in autumn was observed. The results are close to the overall seasonal birth pattern presented by Darrow et al. (2009). According to the authors, birth rates peaked in late summer–early autumn and declined in April–June and November–January (Darrow et al. 2009). The established tendency in our study differs partially from the results reported by Bobak and Gjonca (2001), who observed most births from March–April while the lowest number of births observed from October to December in the Czech Republic.

Weight and length at birth are important features for the growth and development of infants, for morbidity and mortality during the prenatal and postnatal periods and adulthood. The fetus grows rapidly from the fourth to sixth month (i.e., 2nd trimester) of the intrauterine period, when organs increase in size and mass and differentiate functionally. Following that, the rate of growth slows by the time of birth (Harrison et al. 1977). The study of seasonal variations in birth sizes is a common method used to determine the factors that influence birth weight (Chodick et al. 2009). According to our results, the variation in mean birth weight and length depends on the season of birth and maternal sunlight exposure during pregnancy. There was no significant annual variability in birth weight in both sexes. The highest values for birth length were observed in winter for both sexes. Spring appears to be the season with the lowest birth length in boys and in girls, it is the summer. We suppose that this may be due to the sesonal variability of sunshine duration and intensity of UV radiation, which are closely related to the fetal vitamin D levels. Sunshine is the longest and radiation is most intense during the summer and early autumn months, and opposite during the winter ones. Our results differ from the results reported by other authors (Selvin and Janerich 1971; Siniarska and Kozieł 2010; Mladenova 2012). For instance, Selvin and Janerich (1971) studied birth weight in infants born in New York State and reported that those born in winter and spring were slightly heavier and longer compared to those born in summer and autumn. The data for Polish newborns showed that boys exhibited the highest values of weight and length in October and the lowest in March. In newborn girls, the weight and length were the greatest in July and August, respectively, and the lowest in April (Siniarska and Kozieł 2010).

Maternal sunshine exposure during pregnancy has a significant positive impact on birth weight, which is different between trimesters. Equally, pregnant women are considered vulnerable to extreme heat waves and cold spells which have been linked to pregnancy complications including preterm birth and low birth weight (Zhang and Wang 2017; Strand et al. 2011; Chersich et al. 2020; Hajdu and Hajdu 2021; Samuels et al.

2022). The second and third trimesters appear to be slightly more sensitive to temperature exposure compared to the first trimester. Infants with low birth weight have a higher incidence of diseases and disabilities, which continues into adulthood. Several studies have reported associations between the month, or season of birth and risks of later life health outcomes (Disanto et al. 2012) such as type 1 diabetes (Kahn et al. 2009) and multiple sclerosis (Willer et al. 2005), cardiovascular disease (Reffelmann et al. 2007), type 2 diabetes (Vaiserman et al. 2009), psychiatric disorders (McGrath et al. 2010; Disanto et al. 2012).

The present results show the impact of seasonal fluctuations and the influence of sunshine duration during pregnancy on newborn body sizes. The two reported years were characterized by a maximum amount of sunlight in summer and a minimum amount in winter. We found some evidence supporting the notion that maternal sunlight exposure during pregnancy is associated with an increment in both weight and length. The sunshine exposure of the pregnant woman influences the maternal 25-hydroxyvitamin D levels (25(OH)D) and reflects on fetal vitamin D levels. An association between seasonal variations in the birth sizes and maternal sunlight exposure during the first and second trimesters of pregnancy is observed, more pronounced in girls. Maternal sunlight exposure during the first and second trimesters of gestation has the greatest impact on the length of spring and autumn-born babies. The exposure to the sun during the third trimester of pregnancy negatively influenced birth sizes, only in girls. This may be related to maximal sunlight in the early months of pregnancy and the levels of vitamin D, which is an important fat-soluble secosteroid for bone development and affect the growth intensity. The sunshine duration and vitamin D status during pregnancy have a significant effect on fetal skeletal development, tooth formation and birth length (Brooke et al. 1981; Waldie et al. 2000). In boys, the association between birth sizes and maternal sunlight exposure during the three trimesters of gestation is weaker. These results are consistent with the relationship reported by Siniarska and Kozieł (2010) regarding the length of boys at birth with the four climatic factors (air temperature, sunlight, humidity, and precipitation) observed during the second trimester of the prenatal period. The authors also found an association between birth weight and the duration of the mother's sun exposure during the second and third trimesters of gestation. The observed relationship between anthropometric features and the variability of sunshine duration during the four seasons is also consistent with data reported by other studies (McGrath et al. 2005, 2010; Chodick et al. 2009; Mladenova 2012). The results presented in this study provide a rather solid evidence of an association between sunshine duration, maternal exposure to sunlight during pregnancy, and newborn sizes. It can be considered indirect evidence for the effect of vitamin D on intrauterine growth and development, which was also documented by other studies (Mannion et al. 2006; Jakubiec-Wisniewska et al. 2022).

Conclusion

In conclusion, our study shows that seasonal fluctuations in the sunshine duration within a year have a significant impact on the length of Bulgarian neonates born in 2000 and 2001. Our results can be helpful in prenatal diagnostics, neonatal care, and health prevention of pregnant women and neonates. A better understanding of the factors influencing variation in birth sizes (weight and length) may provide a new insight into prenatal growth and development and its effect on individuals' health, longevity, and final height.

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Conflict of interests

No potential conflict of interests was reported by the authors.

Ethical Approval

Ethical approval was given by the Ethics Committee of the Institute of Experimental Morphology, Pathology, and Anthropology with Museum – Bulgarian Academy of Sciences (Protocol 15/01.03.2022) and was conducted in agreement with the principles stated in the Declaration of Helsinki for human studies (2008).

Authors' contributions

AD and IYP design the study; IYP, YZ and BK collected the data; AD, IYP, RG and RS oversaw the statistical analysis/ interpretation; AD, IYP, YZ, ZM and AR were the authors of the written content; All authors agree to be accontable for all aspect of the work.

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Association between smoking status and body composition parameters in a young adult population

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ABSTRACT: The purpose of this study was to analyze the association between smoking status and body composition parameters in 19–30 years old slovak population (mean age: 22,38 ± 2,34 years). The sample consisted of 379 individuals, including 143 men and 236 women. Body composition parameters were obtained using segmentation bioimpedance analysis. The results of our study showed that regular smokers had significantly higher values of waist circumference (p = 0.050), body mass index (p = 0.042), waist-to-height ratio (p = 0.027), fat mass index (p = 0.014) fat mass (p < 0.017), pecentual body fat (p = 0.008), trunk fat mass (FM, p = 0.008), leg fat mass (p = 0.029), and visceral fat area (p = 0.017) compared to non-smokers. Using correlation analysis, we detected an increase in FM (kg) values along with the frequency of smoking (r = 0,136; p = 0,009). Moreover, smoking positively correlated with coffee (r = 0.147; p = 0.002), energy drinks (r = 0.259; p < 0.001), and alcohol consumption (r = 0.101; p = 0.035). Smokers also added salt to their food more often (r = 0.132; p = 0.005) and worked less (r = -0.111; p = 0.025). In this study we confirmed the significant association of smoking with the body composition components, while it is responsible for higher adiposity in young adults.

KEY WORDS: fat mass, lifestyle, early adulthood, smoking.

Original article

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Introduction

The tobacco epidemic is one of the biggest public health threats in the world, killing more than 8 million people a year around the world (St Claire et al. 2020). Of these, more than 7 million deaths are attributed to direct tobacco use, while around 1.2 million deaths are caused by non-smokers being exposed to passive smoking (WHO 2021; Perez-Warnisher, Carballosa de Miguel and Seijo 2019; GBD 2019; Tobacco Collaborators 2021). In 2020, the global prevalence of smoking among adults was 32.6% for men and 6.5% for women (Dai, Gakidou and Lopez 2022). Despite the implementation of tobacco control policies that have led to a decrease in smoking prevalence in Europe (EU) over the past two decades, the number of smokers remains high, with 26% of the overall population and 29.0% of young Europeans aged 15–24 years being smokers (Gravely et al. 2017; Feliu et al. 2019; Teshima, Laverty and Filippidis 2022). Smoking-related mortality in the EU is higher (16.0% among adults aged 30 and over) than the global average of 12.0% (Zafar 2014; Janssen, El Gewily and Bardoutsos 2021). Moreover, the COVID-19 pandemic and the stress resulting from the economic crises have been identified as strong factors associated with an increase in smoking in recent years (Ghadban et al. 2022). In Slovakia, the overall smoking prevalence has increased since 2006, with 32.3% of the adult population being currently smoking. Smoking prevalence is higher among men (38.6%) compared to women (26.0%) (WHO 2019).

Cigarette smoking affects the body through the inhalation of carcinogenic substances, such as dusts and tar, as well as the consumption of nicotine and psychoactive chemical in tobacco that leads to dependence (West 2017). Smoking, especially when combined with unhealthy dietary patterns, increases the risk of chronic diseases (Haffner and Taegtmeyer 2003; Saha et al. 2007). Smoking, even on an occasional basis, significantly raises the risk of cardiovascular diseases. Smokers are two to four times more likely to develop coronary heart disease and twice as likely to experience a stroke compared to non-smokers. The risk of cardiovascular damage is influenced by the duration of smoking and the type and quantity of tobacco products consumed (Conklin et al. 2019). While the exact mechanisms of cardiovascular damage are not fully understood, the detrimental effects of smoking on endothelial function are well recognized. Smoking triggers oxidative processes and adversely affects platelet function, fibrinolysis, inflammation, and vasomotor function. These proatherogenic effects double the 10-year risk of fatal events in smokers compared to non-smokers (Gallucci et al. 2020). Futhermore, tobacco smoking is the major preventable cause of cancer in multiple organs. Despite the longstanding decline in smoking prevalence, lung cancer remains one of the most frequently diagnosed cancers in both sexes (Kulhánová et al. 2020). Several studies have also demonstrated the effects of tobacco smoking on the skeletal system. Specifically, recent evidence indicates that smoking disrupts the mechanisms of bone turnover, leading to lower bone mass and bone mineral density (BMD), which increases the vulnerability of bones to osteoporosis (Cusano 2015; Al-Bashaireh et al. 2018) and fractures (Vestergaard and

Mosekilde 2003; Cusano 2015; Zhang et al. 2022). Moreover, cigarettes are assumed to be a risk factor in sleep disorders, including breathing sleep disorders (Witek and Lipowicz 2021).

Studies regarding the effects of smoking on body composition are conflicting. Some studies have found that smokers have a lower Body mass index (BMI) compared to non-smokers, which may be attributed to the metabolic properties of nicotine, leading to increased oxidation and reduced fat accumulation at higher levels (Bamia et al. 2004; Akbartabartoori, Lean and Hankey 2005). However, regular smoking has been found to be associated with higher BMI (Dare, Mackay and Pell 2015; Pruszkowska-Przybylska et al. 2016). Heavy smoking has an impact on fat distribution and is linked to abdominal obesity and insulin resistance (Chiolero et al. 2008). Additionally, unhealthy lifestyle behaviors are often prevalent among heavy smokers, potentially contributing to weight gain and fat accumulation. Pisinger, Toft and Jørgensen (2009) demonstrated a positive correlation between cigarette consumption and an unhealthy diet, higher energy intake, sedentary lifestyle, and alcohol consumption. Cigarette smoke components play a significant role in reducing muscle mass, reducing the amount of oxygen supplied to muscles, and disrupting mitochondrial function. Studies indicate that smokers have lower fat-free mass compared to non-smokers. Substances present in cigarette smoke can stimulate muscle protein breakdown and disrupt protein synthesis. Aldehydes in cigarette smoke enter the circulation and directly impact skeletal muscle by reducing protein synthesis in human muscle cells, inducing muscle atrophy and myosin breakdown (Montes de Oca et al. 2008; Wüst et al. 2008; van den Borst et al. 2011; Kok, Hoekstra and Twisk 2012; Degens, Gayan-Ramirez and van Hees 2015).

The aim of our pilot cross-sectional study is to establish the relationships between smoking status and anthropometric characteristics, obesity indices and body composition parameters in Slovak young adults.

Material and methods

The sample comprised 379 Slovak young adults ranging in age from 19 to 30 years (with mean age 22.38 ± 2.34 y), who were enrolled in this cross-sectional research during 2019–2020. All study participants were evaluated in the Biomedical laboratory of Department of Anthropology at Comenius University during the morning. Women and men were approached and recruited using a nonrandom procedure (based on volunteering and convenience). Each participant provided written informed consent for this study which adhered to the Declaration of Helsinki principles. Biomedical research was also approved by the ethics committee of the Faculty of Natural Sciences at Comenius University with number ECH19021.

Data on lifestyle were collected from participants using a questionnaire (a modified version of the STEPS questionnaire WHO v.3.2), which was part of a study designed for research purposes. Probands were asked to complete a questionnaire containing questions related to their socio-demographics background and lifestyle. All socio-demographic and lifestyle variables were measured by self-reporting. Smoking status was categorised as regular (smoking once a week to every day) and non-smokers (never smoking).

Anthropometric measurements were taken using standard anthropometric techniques by trained anthropologists. Body height was measured with participants standing without shoes and heavy outer garments by anthropometer. Waist and hip circumferences were measured to the nearest 0.5 cm using a non-elastic tape. Waist circumference (WC) was measured at the level of the umbilicus, and the hip circumference (HC) was measured at the maximum posterior protrusion of the buttocks. BMI was calculated as body weight in kilograms divided by height squared (WHO 2000) and values below 24.9 kg/m^2 were considered as optimal. Waist-to-hip ratio (WHR) was calculated as the circumference of the waist divided by the circumference of the hips (WHO 2000) and values less than 0.84 for women and less than 0.89 for men were considered as optimal. Waist-to-height ratio (WHtR) was calculated as the circumference of the waist divided by height squared (Schneider et al. 2010) and values below 0.48 for women and less than 0.52 for men were considered as optimal. The InBody 770 body composition analyzer (Biospace Co., Korea) was used to detect the human body composition parameters based on the recommendation provided in the user manual. Participants were tested in the quiet state in the morning. Participants stood barefoot on the pedal plate electrode. The hands were naturally hanging and held the hand electrode gently, keeping the angle at 15° between the torso and the upper limbs. The test included measurements of body weight, lean body mass (LBM), intracellular fluid (intacellular water, ICW), extracellular fluid (extracellular water, ECW), body water content (total body water, TBW), skeletal muscle (skeletal muscle mass, SMM), body fat (FM) and body cell mass (BCM). Fat mass index (FMI) was calculated as FM divided by height squared (kg/m^2). Visceral Fat Area (VFA) is based on the estimated amount of fat surrounding internal organs in the abdomen, and values below 100 cm^2 were considered optimal. These detailed body composition variables were displayed on a computer using the LookinBody programme software.

All statistical analyses were performed with IBM SPSS for Windows (Statistical Package for the Social Science, version 25.0, Chicago, IL) and statistical significance was defined as $p \leq 0.05$. The assumption of normal distribution was tested by the one-sample Kolmogorov-Smirnov test. We used covariate-adjusted generalized linear models (GLMs) to analyze the effects of smoking (no = 0, and yes = 1) on body composition parameters (as dependent variables), with age and gender as the covariates. Correlation analysis was used to test the strenght of association between two variables and the direction of the relationship. To assess correlation between smoking frequency and body composition parameters the Pearson's chi-square test was used in case of normally distributed variable and Spearman nonparametric test for data with non-normal distribution. Kendall's tau-b correlation coefficient was used to measure strength and direction of association between two variables measured on at least an ordinal scale. This type of correlation was used to correlate the mutual relationships of lifestyle factors.

Results

Characteristics of the study participants, anthropometric characteristics and obesity indices according to smoking status in young adults

The sample consisted of 379 individuals – 143 men (37.7%) and 236 women (62.3%). Among the participants, 12.8% were regular smokers, while 87.2% were non-smokers. The prevalence rate of alcohol consumption was 33.3%. Anthropometric characteristics, obesity indices according to smoking status in young adults are summarized in Table 1. After adjustment for age and gender, we found statistically significant differences between regular smokers and non-smokers. Regular smokers attained significantly higher values of waist circumference than non-smokers (smokers: 78.24 ± 9.97 cm vs. non-smokers: 74.22 ± 9.89 cm; $p = 0.050$). Additionally, results indicated that regular smokers had statistically significantly higher BMI ($p = 0.042$), WHtR index ($p = 0.027$) and FMI ($p = 0.014$).

Table 1. Anthropometric characteristic and obesity indices according to smoking status in young adults

Number of participants	*Regular smokers*			Non-smokers			p^*
	N = 47			*N = 320*			
	Mean		SD	Mean		SD	
Height, cm	172.63	±	9.74	171.83	±	9.17	0.423
Weight, kg	72.89	±	15.03	67.72	±	14.79	0.130
Waist circumference, cm	78.24	±	9.97	74.22	±	9.89	0.050
Hip circumference, cm	99.53	±	8.91	96.89	±	8.41	0.131
BMI, kg/m^2	24.32	±	3.85	22.79	±	3.89	0.042
WHR	0.79	±	0.08	0.76	±	0.06	0.104
WHtR	0.45	±	0.05	0.43	±	0.05	0.027
FMI, kg/m^2	6.45	±	2.70	5.61	±	2.80	0.014

Note: N, number of participants; SD, standard deviation; p*, value of statistical significance adjusted for age and sex; BMI, body mass index;

WHR, Waist-to-hip ratio; WHtR, Waist-to-height ratio; FMI, Fat mass index

Body composition characteristics according to smoking status in young adults

The mean values of bioelectric impedance variables are shown in Table 2. After adjustment for age and gender, statistically significant differences were observed between regular smokers and non-smokers in terms of body composition parameters. Regular smokers had higher mean values of FM (smokers: 19.01 ± 7.47 kg vs. non-smokers: 16.36 ± 7.84 kg; $p < 0.017$), PBF (smokers: 25.81 ± 8.29 % vs. non-smokers: 24.02 ± 8.48 %; $p = 0.008$), trunk FM (smokers: 9.70 ± 4.01 kg vs. non-smokers: 8.06 ± 4.03 kg; $p = 0.008$), leg FM (smokers: 2.85 ± 1.07 kg vs. non-smokers: 2.55 ± 1.08 kg; $p = 0.029$), and VFA (smokers: 83.16 ± 37.38 cm^2 vs. non-smokers: 70.56 ± 38.93 cm^2; $p = 0.017$) compared to non-smokers.

Table 2. Body composition characteristics according to smoking status in young adults

Number of participants	*Regular smokers* N = 47			Non-smokers *N = 320*			p^*
	Mean		SD	Mean		SD	
Skeletal Muscle Mass, kg	30.15	±	7.28	28.54	±	7.49	0.964
Fat Free Mass, kg	53.47	±	11.81	51.36	±	12.29	0.590
Lean Body Mass, kg	50.71	±	11.29	48.32	±	11.60	0.896
Lean Body Mass Right Arm, kg	2.88	±	0.88	2.66	±	0.90	0.733
Lean Body Mass Trunk, kg	23.77	±	5.26	22.38	±	5.36	0.600
Lean Body Mass Right Leg, kg	8.45	±	1.87	8.19	±	1.94	0.400
Fat Mass, kg	19.01	±	7.47	16.36	±	7.84	0.017
Percentual Body Fat, %	25.81	±	8.29	24.02	±	8.48	0.008
Visceral Fat Area, cm^2	83.16	±	37.38	70.56	±	38.93	0.017
Fat Mass Right Arm, kg	1.23	±	0.67	1.06	±	0.81	0.080
Fat Mass Right Leg, kg	2.85	±	1.07	2.55	±	1.08	0.029
Fat Mass Trunk, kg	9.70	±	4.01	8.06	±	4.03	0.008
Total Body Water, l	39.43	±	8.72	37.59	±	8.97	0.888
Intracellular Water, l	24.65	±	5.58	23.52	±	6.09	0.761
Extracellular Water, l	14.78	±	3.15	14.17	±	3.24	0.776
Body Cell Mass, kg	35.30	±	8.00	33.55	±	8.23	0.956

Note: N, number of participants; SD, standard deviation; p*, value of statistical significance adjusted for age and sex

Correlation analysis between smoking frequency and body composition parameters

Subsequently, the participants were divided into six groups based on smoking frequency: non-smokers (n = 308), smoking less than once a month (n = 12), smoking one to two days a week (n = 10), smoking three to four days a week (n = 9), smoking five to six days a week (n = 4), and smoking daily (n = 24). We investigated how smoking frequency correlateed with body composition parameters. Statistically significant correlations are shown in Table 3. The positive correlation coefficients indicate that all adiposity parameters increases with the higher frequency of smoking.

Correlation analysis between smoking status and selected lifestyle factors

By performing a correlation analysis, we found that smokers had a higher frequency of coffee consumption (r = 0.147; *p* = 0.002), energy drinks consumption (r = 0.259; *p* < 0.001), and alcohol drinking (r = 0.101; *p* = 0.035) compared to non-smokers. Smokers also added salt to their food more often (r = 0.132; *p* = 0.005) and worked less (r = -0.111; p = 0.025; data not shown in table).

Table 3. Correlation analysis between smoking frequency and body composition parameters

	r	*p*
Waist circumference, cm	0.131	0.012
Hip circumference, cm	0.124	0.018
BMI, kg/m^2	0.134	0.010
WHR	0.105	0.044
WHtR	0.158	0.002
FMI, kg/m^2	0.121	0.021
Fat Mass, kg	0.136	0.009
Fat Mass Trunk, kg	0.151	0.004
Visceral Fat Area, cm^2	0.135	0.009

Note: r, correlation coefficient; p, value of statistical significance; BMI, body mass index; WHR, Waist-to-hip ratio; WHtR, Waist-to-height ratio; FMI, Fat mass index

Discussion

In our study, we recorded higher values of all anthropometric parameters and indexes in smoking young adults than in non-smokers, with statistically significant differences in waist circumference, BMI, WHR, WHtR and FMI indexes. Studies examining the effect of smoking on anthropometric parameters are contradictory. Some previous studies have shown negative correlation between obesity and smoking. For example, in an older study of Akbartabartoori et al. (2005) on Scottish adolescents and adults aged 16 to 74 years, statistically significantly lower average values of BMI and hip circumference were recorded in the smokers group (BMI men = 25.6 kg/m2, BMI women: 25.2 kg/m2; hips men: 101.2 cm, hips women = 100.5 cm) compared to non-smokers (BMI men = 26.8 kg/m2, BMI women = 26.2 kg/m2; hips men = 101.9 cm, hips women = 101.9 cm). In our study, we observed statistically significantly higher BMI values in smokers (24.32 ± 3.85) than in non-smokers (22.79 ± 3.89). Also, hip circumference was higher in smokers (99.53 ± 8.91 cm) than in non-smokers (96.89 ± 8.41 cm), but the difference was not statistically significant. Assyov et al. (2008) found that smoking was associated with WC, while smokers had higher values than non-smokers (105.5 ± 7.84 cm vs. 99.73 ± 9.65cm), which was consistent with the results of our study (smokers: 78.24 ± 9.97 cm; non-smokers: 74.22 ± 9.89 cm). Chiolero et al. (2007) in their study of the Swiss population aged 25 to 44 years recorded a lower average BMI value in non-smoking men (24.6 ± 0.1) than in heavy smokers (24.9 ± 0.2), but higher than in light smokers (24.1 ± 0.2) and the same as in moderate smokers (24.6 ± 0.2). Smokers were defined based on the number of cigarettes smoked per day. Light smokers were defined as those smoking one to nine cigarettes per day, moderate smokers smoked ten to 19 cigarettes, and heavy smokers smoked 20 or more cigarettes per day. Also, Dare et al. (2015) found out that among smokers, the risk of obesity increased with the quantity of cigarettes smoked and former heavy smokers were more likely to be obese compared to former light smokers (adjusted OR 1.60, 95% 1.56 – 1.64, $p < 0.001$). Additionally, Pruszkowska-Przybylska et al. (2016) revealed a significant influence of nicotinism on BMI: regular cigarette smoking was associated with higher BMI values in students aged between 19 and 26. The different results of smoking studies may indicate that endogenous factors and other lifestyle factors may be more important in influencing body composition than the direct effects of smoking (Efendi et al. 2018).

In terms of body composition parameters, statistically significant differences were observed in the case of FM (kg) as well as PBF (%). Significantly higher FM values were recorded in all compared body segments except for the arm (kg) FM. Smokers had a higher visceral fat area (83.16 ± 37.38 cm^2) compared to non-smokers (70.56 ± 38.93 cm^2). Correlation analysis confirmed a positive relationship between FM (kg) and smoking frequency. Assyov et al. (2008) obtained similar results in their study on the effect of smoking on body composition, where smokers had higher values of FM (36.62 ± 8.46 %) than non-smokers (32.66 ± 6.96 %). Stefan et al. (2017) examined the influence of lifestyle factors and body composition on Croatian adults (with a mean age of 19.81 years). The correlation analysis showed that FM (%) increased in both male and female smokers, with a statistically significant correlation observed only in females (r = 0.230; p < 0.010). In a study by Alkeilani et al. (2022)

on adults aged 18 to 89 years from the population in Qatar, smokers had higher mean FM values (29.0 ± 11.3 kg) compared to never smokers (23.4 ± 6.6 kg, $p < 0.001$), which was consistent with the findings of our study.

In our study we found that smokers had a higher frequency of coffee consumption, energy drinks, and alcohol compared to non-smokers. This unhealthy lifestyle could contribute to an increased BMI and FM observed among smokers in our study. Furthermore, we noticed a decrease in the number of worked participants among smokers, which could lead to lower energy expenditure and consequently an increase in FM values.

Previous studies have also reported higher caffeine intake in smokers compared to non-smokers in the general population (Hewlett and Smith 2006; Mahoney et al. 2019). Results from the Riera-Sampol et al.´study (2022) showed that the association between smoking and caffeine consumption may depend on the caffeine source. They found a higher prevalence of coffee, cola and energy drink consumption among smokers, while no differences were observed for other caffeine sources considered. Physiological, cognitive, and environmental factors may all contribute to the association between smoking and caffeine intake, because smoking increases the rate of caffeine metabolism, as a consequence, smokers must consume caffeine more frequently than non-smokers to maintain similar effects (Bjørngaard et al. 2017). Pisinger, Toft, and Jørgensen (2009) also recorded significantly higher consumption of unhealthy food, alcohol, overall energy intake, and a sedentary lifestyle among smokers ($p < 0.001$).

Despite our interesting findings, there are some limitations that need to be acknowledged. The study is cross-sectional and may have had selection bias during participation recruitment, and the particular study design may limit result generalization to all Slovak young adults. Since data collection was done using a self-answered questionnaire, some of the participants might not have been comfortable with reporting their true smoking status. Moreover, the study population was smaller (n = 379) and for future studies, it would be paramount to enlarge and divided by gender the study sample for a detailed analysis.

Conclusion

In this study, we have provided novel data that supports the significant association of smoking status and body composition parameters in Slovak young adults. The findings of this study may have important implications for public health, as they could help to inform interventions aimed at reducing smoking rates and promoting healthy body composition in young adults.

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Conflict of interest

The authors declare that there is no conflict of interest.

Authors' contribution

DF participated in collection of data, analysis and interpretation of data, and the writing of the manuscript. LV participated in collection of data, analysis and interpretation of data, and the writing of the manuscript. RB was responsible for the statistical analysis.

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Where are the Roman women of Ovilava? A spatio-temporal approach to interpret the female deficit at the eastern Roman cemetery (Gräberfeld Ost) of Ovilava, Austria

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Abstract: In historic and prehistoric populations, the sex ratio can often only be determined based on bioarchaeological analyses of cemeteries. In this context, a so-called female deficit has been described, which clearly contrasts the sex ratio typical of recent populations. The present study aims to analyze the sex ratio in the eastern cemetery ('Gräberfeld Ost') of the Roman town Ovilava. This site is located beneath today's city of Wels in Upper Austria (AUT), in the northwestern region of Noricum. Spatial as well as chronological aspects of the sex ratio were considered. A total of 111 individuals older than 15 years could be included in the analysis. Radiocarbon dating allowed a chronological classification of the burials. In addition, a spatial analysis of the cemetery was carried out using a geoinformation system. For the whole sample, a sex ratio of 200.0 and a marked female deficit were recorded. Considering the individual age classes and the periods separately, an apparent female surplus emerged among individuals younger than 20 years (sex ratio = 62.5). With increasing age class, the proportion of females reduced dramatically; among the over 40-year-olds, the sex ratio was 370.0. There was also an apparent female deficit in the Early Roman and Imperial Roman Periods but an almost balanced sex ratio in the early Middle Ages. Furthermore, the sex proportion varied between the cemetery areas. Consequently, a female deficit occurs mainly during Roman times and in specific areas of the burial ground.

Key words: female deficit, Roman burials, sex ratio, Noricum, bioarchaeology.

Original article

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Introduction

In 1662, the English businessman John Graunt (1620–1674) published the first detailed descriptive analysis of the numerical ratio of male to female newborns. His analysis of the sex ratio of baptized children in London revealed a significantly higher number of male newborns than female ones (Greuch 2020). This observation of a numerical preponderance of male infants was confirmed by both the English mathematician John Arbuthnott (1667–1735) and the German demographer Johann Süßmilch (1707–1767) (Greuch 2020). On the other hand, Charles Darwin (1809–1882) postulated a 1:1 ratio of male to female individuals in a natural population (Darwin 1871). This assumption was later adopted by Ronald Fisher (1890–1962), whose theory of a male-to-female ratio of 1:1 became known as Fisher's principle as an evolutionarily stable strategy (Greuch 2020). All these observations are related to the so-called secondary sex ratio, the numerical sex ratio at the time of birth or the neonatal period (Chao et al. 2019; DÁlfonso et al. 2023). In addition to the secondary sex ratio, the primary sex ratio is also defined as the numerical ratio of chromosomally female (XX constellation) to chromosomally male (XY constellation) zygotes at the time of conception. However, this primary sex ratio cannot be examined in demographic analyses (James 1987).

Currently, in populations without prenatal selection favoring one sex, a secondary sex ratio of 103–105 male newborns per 100 female newborns is observed (Hobbs 2004). However, a marked female deficit is observable in populations with prenatal selection against female offspring (Sharma et al. 2017). Following the neonatal period, which concludes on the 42nd day post-birth, the tertiary sex ratio becomes established. As individuals age, this ratio gradually shifts in favor of the biological female sex. Throughout adolescence and in the majority of human populations, males tend to exhibit higher mortality rates compared to females within the same age group. This discrepancy persists even after accounting for causes of death specific to females, such as childbirth-related fatalities. The higher mortality among males can be attributed to both natural factors, including heart attacks, strokes, and cancer (which account for the majority of deaths), as well as violent causes like homicide and warfare (Kruger and Nesse 2004). Consequently, females generally enjoy a longer life expectancy due to these factors. The low status of the female gender, however, increases female mortality rates and contributes in this way to an imbalance between the sexes and may lead to a female deficit, even today (Coale 1991). Consequently, the sex ratio (SR) varies considerably around the world, from a dramatic male surplus in states such as Qatar (SR=266), the United Arabian Emirates (SR= 228.2) or Bahrain (SR = 163.8) to an apparent female surplus, as in the successor states of the former Soviet Union such as Georgia (SR= 88.7), Lituania (SR=88.4), Russia (SR=86.7), Lativa (SR=86.7), Ukraine (86.2), Belarus (SR=85.5) and Armenia (SR=81.9) (UNO 2022). These differences can be biological-medical but also ecological, sociocultural, political-historical, or economic (Heshketh and Xing 2006).

A particular challenge is the reconstruction of the sex ratio of historical populations. On the one hand, written sources can be used; on the other hand, if written sources are unavailable, bioarchaeological analyses of cemeteries are the only way to reconstruct the sex ratio among historical and prehistorical populations. In the case of bioarchaeological, i.e., osteological analyses, only the tertiary sex ratio of juvenile or adult individuals can be determined with macroscopic-morphological methods since sex-typical features on the skeleton only develop with the hormonal changes of puberty (Schaefer et al. 2009). Sometimes, however, a purely morphological sex diagnosis is impossible due to a poor state of preservation or an indifferent combination of features (White and Folkens 2005). Although there are limits to osteological analysis, bioarchaeological analyses often reveal an astonishing phenomenon that stands in marked contrast to observations of recent populations:

In numerous studies, a conspicuous surplus of male-classified adult individuals was reported (Schott 1964; Häussler 1969; Weiss 1972; Caselitz 1981; Wittwer-Backofen 1987; Volk and Büchner 1995), a finding that led to an assumption – rarely critically questioned – of a general female deficit in archaeological series (Kemkes-Grottenthaler 1997). This situation led to the point that if no female deficit was found, this finding was interpreted as a methodological artifact (Schott 1964; Kunter 1991). The female deficit in bioarchaeological series remained a mystery, although more than 25 years ago, Ariane Kemkes-Grottenthaler (1997) presented a critical analysis of possible explanations of this phenomenon. She discussed the importance of methodological problems in diagnosing sex on the skeleton, the focus on selected samples such as monastery cemeteries, poorhouses, or slave cemeteries (Weiss 1972; Caselitz 1981), but also sex-typically varying mortality patterns, or the theory of sex-differentiated parental investment according to Trivers and Willard (1973).

In recent years, however, descriptions of a general female deficit have been increasingly critically questioned, and various explanatory models have been postulated. Mostly, methodological problems in morphological sex estimation resulting in the benefit of male classifications are reported (Weiss 1972). White and Folkens (2005) discussed artifacts, such as misclassification by using only skull elements for sex determination and the problem of sex determination among juvenile individuals. Other studies yielded no significant differences in the preservation between male and female skeletons (Stojankowski et al. 2002; Walker et al. 1988). Cintas-Peña and Herrero-Corral (2020), who reported the apparent male surplus at 62 prehistoric sites on the Iberian Peninsula from the 8th to the 3rd millennium BC, discussed – but finally rejected the interpretation that a possibly poor state of preservation of female skeletal remains or the complete destruction of the smaller and more delicate female bones, might lead to the impossibility of a correct sex diagnosis.

However, a male surplus is not only described for prehistoric cemeteries; early and late medieval samples from Italy (Barbiera et al. 2017) and medieval England (Bardsley 2014) also suggest a clear male surplus. In contrast, a balanced sex ratio is described for Late Antique Italy (Barbiera et al. 2017). Again, this finding contrasts with an apparent male surplus or female deficit described for numerous Roman burial sites in Austria (Lebzelter 1935; Ehgartner 1947; Kloiber 1957, 1962; Pichlmayer 1972; Neugebauer 1987; Wiltschke and

Teschler-Nicola 1991; Berner 2014; Marschler 2013), Germany (Ziegelmayer 1979; Wahl 1988), and Switzerland (Brunner 1972), but also in the former Yugoslavia (Mikic 1984). A female surplus, on the other hand, is documented much more rarely (Pail 2009; Schweder and Winkler 2004). These very unbalanced gender relations of the Roman period skeletal series, however, are rarely subjected to a critical analysis. Therefore, the present study aimed to analyze and interpret the sex ratio of the Roman and early medieval inhumations at the eastern cemetery ('Gräberfeld Ost') of Ovilava, now known as Wels, in present-day Upper Austria (AUT). For the first time at this crucial urban Roman site, we applied a state-of-the-art archaeothanatological approach. This attempt integrates bioarchaeological, spatial archaeological, and social archaeological methods. Since the analysis of the sex ratio is based on the morphological analysis of the skeletal features, only the biological sex is considered in the present study.

For this purpose, the following hypotheses were tested:

1. The cemetery of Ovilava shows a female deficit.
2. The sex ratio of the cemetery of Ovilava varies depending on the burials' chronology and the burial areas' spatial patterns.

Material and methods

The site Ovilava – archaeological background

For around 500 years, between the ends of the 1st cent. BC and the 5th cent. AD, the southeastern Upper Danube River Basin (sUDRB) (Fehér et al. 2015; Sommerwerk et al. 2022; Stagl and Hattermann 2015; Zoboli et al. 2015), comprising roughly the Roman provinces of Noricum and Pannonia, was under Roman rule and part of the *Imperium Romanum* (Hagmann 2019). Systematic civil settlement activities in Noricum emerged traceable from the 2nd half of the 1st cent. AD onwards, along the border zone next to the Danube river, known today as the UNESCO World Heritage site 'Danube Limes,' but called '*ripa Norica*' in ancient times (Hagmann 2019, 2022, 2023). The Roman town of *Ovilava/Ovilavis* (coordinates: 48.1575°, 14.026667° [EPSG:4326 WGS 84; https://epsg.io/4326]), which today can be found in the urban area of the city of Wels in Upper Austria (AUT), was located in the northwestern part of Roman *Noricum* (resp., in late antiquity, *Noricum ripense*), around 20 km south of the river Danube. Throughout hundreds of years of Roman settlement, Ovilava never served as a military camp near the Danube limes. Instead, it consistently functioned as a civilian and economic center in the hinterland of the frontier zone. Consequently, the inhabitants of Ovilava were mostly not active soldiers but mainly civil traders, craftsmen, and other kinds of workers, which likely originated from Norcium and adjacent regions. Nevertheless, some may also be migrated from other parts of the Roman Empire, such as North Africa, to this area near the Danube *limes* (Antonio et al. 2022) (Figure 1).

Currently, there is no archaeological evidence of any older, i.e., pre-Roman, settlement in this region. The first traces of a Roman settlement preceding the urban place can be assumed in the 2nd half of the 1st cent. AD and can be possibly thought of as a rural *vicus* (roadside settlement). In the area of Ovilava, the east-west running trunk road that connected, e.g., in the 2nd cent. AD, the Roman town (*municipium*) and legionary fortress (*castra legionis*) *Lauriacum* (today Enns, AUT) and the town (*municipium*) of *Iuvavum* (today Salzburg, AUT), as well as the road that led from the supra-regional trading hub Aquilea in today's northern Italy over the Alps to the military camp (*castellum*) of Boiodurum (today Passau, GER), crossed each other. The favorable location at the intersection of two critical roads suggests that Roman Ovilava established itself as an important administrative, infrastructural, and economic node in the province of Noricum. At the beginning of the 2nd cent. AD, when the whole region was at its height, Ovilava received Roman city rights during the reign of the Roman emperor Hadrian (76–138 AD) and became *municipium Aelium Ovilava*. During the second half of the 2nd cent. AD Ovilava seemingly remained untroubled by the Marcomannic wars. Ovilava was finally raised to *Colonia Aurelia Antoniniana* during the reign of Caracalla (211–217 AD), and town fortification walls were erected during the 3rd cent. AD. During the late-antique administrative reforms, e.g., by Diocletian (284–305 AD), Ovilava probably became the capital of the new province of *Noricum ripense*. (Miglbauer 2002, 2014, 2015)

Fig. 1. Spatial location and extent of the Roman town of Ovilava and the eastern cemetery ('Gräberfeld Ost') in former northwestern Noricum, situated within the area of modern-day Wels in Upper Austria (AUT)

A long-lasting polycrisis, resulting from epidemics like the 'Antonine plague' in the late 2nd cent. AD or the 'plague of Cyprian' in the mid-3rd cent. AD, climatic changes with the end of the Roman Climate Optimum (RCO) and the beginning of the Late Antique Little Ice Age (LALIA) (Erdkamp 2021; Haldon et al. 2018b, 2018a, 2018c; Harper 2017; McCormick et al. 2012), various migration waves in the course of the great migration period (Meier 2019), as well as the regular massive internal political conflicts in the style of civil wars as also conflicts with external enemies (Börm 2022), triggered the relatively slow but somewhen ultimate decline of northern Noricum as an integral part of the Roman Empire (Hagmann 2019, 2022, 2023).

In 476 AD, the barbarian king Odoacer deposed Romulus August(ul)us, who is often considered the last emperor of the Western Roman Empire. Odoacer replaced him as the new king of the Italian peninsula's lands, including Noricum. According to the *Vita sancti Severini* (44, 5), the life description of *Severinus* of Noricum (c. 410–482 AD), finished in 511 AD by the monk Eugippius and the most important written source for the 'latest' late-antique period of *Noricum ripense*, Odoacer's brother Hunuwulf followed the king's order to resettle the Roman people (officials as well as the 'average' population) from Noricum in 488 AD to the South (Winckler 2016). It cannot be verified that the entire population left the region because of that order and also not how the whole process took place (non-violent? forced?) but, e.g., the eastern cemetery of Ovilava (cf. *infra*) was only seldom used from the end of the 3rd cent. AD onwards. It was reoccupied again by early medieval populations, probably Bavarians, only in the early 7th cent AD. Therefore, Roman settlement activities seemingly ended in the 5th cent. AD to a likely considerable extent, although it hardly came to a settlement hiatus (Hausmair 2022). It is thus not clear what really happened to the Roman population of this area.

The eastern burial ground of Ovilava – the 'Gräberfeld Ost'

According to Roman custom, the civil inhabitants of Ovilava were buried in the necropoleis along the arterial roads outside the city gates (Retief and Cilliers 2010, 134). One of the town's most significant cemeteries was located in the east along the road from *Lauriacum* to *Iuvavum* and is accordingly known as 'eastern cemetery' ('Gräberfeld Ost'). Today the area of the Roman cemeteries is located in the densely built city of Wels. Therefore, excavations are only possible in the context of construction projects, mainly within the modern city – recorded archaeological finds are thus known since the 19th cent. Until today, it was not possible to uncover the entire burial ground, but only several unconnected areas have been excavated. (Jäger-Wersonig 1999a,b,c; Miglbauer 2007, 2009) For the study presented in this paper, 5 specific excavation areas from the Gräberfeld Ost are of utmost importance:

– Pendlerparkplatz (2004–2005) (Miglbauer 2006; Greisinger 2006; Greisinger et al. 2020);
– Kanalgrabung Dr.-Groß-Straße (2006) (Greisinger 2007; Greisinger et al. 2020);
– Alois-Auer-Straße (2007) (Miglbauer 2006; Greisinger et al. 2020);
– Dr.-Groß-/Dr.-Schauer-Straße (2002) (Miglbauer 2004; Greisinger et al. 2020);
– Fronius (2008–2010) (Greisinger 2010; Greisinger et al. 2020).

The construction of an underground car park near the central train station resulted in a rescue excavation of large parts of the Roman cemetery between 2004 and 2005 ('Pendlerparkplatz'). During this campaign, more than 220 burials (61 inhumations and more than 150 cremations) could be excavated (Greisinger 2006). The construction of a drainage channel in Dr.-Groß-Straße ('Kanalgrabung') in 2006 also required a rescue excavation, during which 16 inhumation graves were uncovered (Greisinger 2007). Between 2002 and 2007, rescue excavations in the Alois-Auer-Straße and Dr.-Groß-Straße respectively Dr.-Schauer-Straße yielded in total the remains of 8 inhumations (Miglbauer 2004, 2006). All these excavations occurred between the city center and the central train station of Wels. Somewhat further outside today's city center in the suburbs east of the city center, 51 inhumations were recovered between 2008 and 2010 in the course of a rescue excavation on the premises of the Fronius company (Greisinger 2010). As pointed out, the cemetery of Ovilava was bi-ritual, containing cremations and inhumations. The different sections of the burial ground treated in this study are presented in Figure 2.

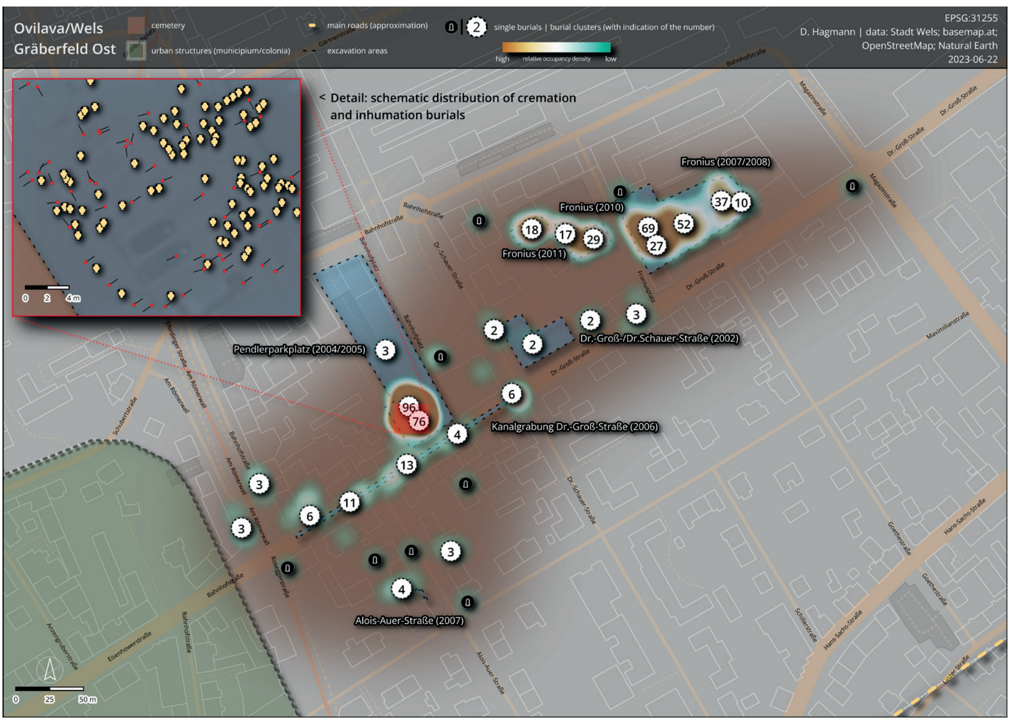

Fig. 2. Spatial extent of the eastern cemetery indicating the 5 excavation areas discussed in this study, including the years of excavation: Pendlerparkplatz (2004–2005), Kanalgrabung Dr.-Groß-Straße (2006), Alois-Auer-Straße (2007), Dr.-Groß-/Dr.-Schauer-Straße (2002), and Fronius (2008–2010). A section of the Pendlerparkplatz area is highlighted in detail and displays the predominantly distinct spatial separation between inhumation burials (stylized bodies, indicating their respective orientation) and cremation burials (urn symbols)

Taking into account the excavations from the 19th and 20th cent. a partial picture of the cemetery east along a *decumanus* (east-west street in the street grid) can be reconstructed in terms of its extent (Miglbauer and Waser 2020). The spatial locations of the excavated cemetery areas in the outskirts of the urban settlement as well as the cemetery itself, the (yet partially analyzed) grave goods, and (few) qualitative attributes of the tombs indicate that elite but also 'commoners' and 'poor' were buried at the burial ground of Ovilava. According to the archaeological finds, the burials dated mainly in the 2nd and 3rd cent. AD (Jäger-Wersonig 1999 20–22; Miglbauer 2007, 2009). Considering previous investigations, the cemetery seemed to be in use, although possibly very scarce in the end, up to Early Medieval times (Miglbauer 2001; Hausmair 2022). Few burials are attributed to Early Medieval Bavarians (Hausmair 2006).

Over 2/3 of the burials consisted of cremations; the remaining others were inhumations. The skeletal remains have been stored at the Archaeological Collection of the Stadtmuseum Wels-Minoriten. In 2019, the skeletons were temporarily transferred to the Department of Evolutionary Anthropology, University of Vienna, where the bioarchaeological analysis is currently in progress. The remaining cremations of the burial ground of Ovilava have not been analyzed up to now and are subject to a planned project which is scheduled to start in 2024.

Chronology

In order to be able to assess the chronological position of the individual cemetery areas better and to be able to reconstruct the settlement history based on archaeothanatological findings, we decided to carry out radiocarbon dating of inhumations of the cemetery as much as possible (Pearce 2016). The radiocarbon dating of 97 individuals, with 83 individuals older than 15 years, has been carried out so far. Following a dedicated sampling strategy, clusters for pre-selection were spatially defined, and individuals from all cemetery areas were selected as a proportion of the total number of burials and dated using radiocarbon analysis. The samples determined in this way were further supplemented by individual samples of further individuals particularly suitable for dating. The radiocarbon datings were carried out at the VERA-lab of the Faculty of Physics at the University of Vienna (https://isotopenphysik.univie.ac.at/en/vera/).

A dedicated controlled vocabulary, explicitly tailored for Noricum, was used to periodize the data (Hagmann 2021): Based on the radiocarbon dating results, the inhumations were classified using the following time periods for Roman Antiquity (50 BC to 500 AD) in Noricum (resp. 50 AD to 500 AD in northern Noricum) as

- Early Period (50 AD to 117 AD);
- Imperial Period (117 to 284 AD);
- Late Antiquity (284 to 488 AD).

Early Middle Ages (for the Gräberfeld Ost, this period corresponds c. to the 7th and 8th cent. AD) were defined from 500 to 1100 AD following Eichert and Richards (2022).

21 individuals belonged to the Early Period, 58 to the Imperial Period, 2 to Late Antiquity, and 16 were classified as Early Medieval.

Sample

The excavated parts of the Ovilava burial ground include 328 cremation burials and 134 inhumations. A total of 153 individuals could be assigned to the inhumations. The spatial distribution of the burials is presented in Table 1.

All 153 individuals were osteologically analyzed using standard bioarchaeological protocols outlined by Buikstra and Ubelaker (1994) as well as Mitchell and Brickley (2017). Age at death and sex of the individuals were estimated by a combination of various standard techniques (White and Folkens 2005). Pathologies were determined according to Ortner (2003). Although the state of preservation of the individual skeletons differed, the preservation of the pelvis and/or the skull of 108 individuals older than 15 years allowed sex as well as age at death estimation.

Age at death determination

Age at death of non-adult individuals was estimated through dental development and eruption patterns, the degree of closure of the epiphyseal plates (Cardoso 2008; Coqueugniot and Weaver 2007; Schaefer et al. 2009), and metric documentation of diaphyseal lengths (AlQathani et al. 2010; Cunningham et al. 2016). If the specific skeletal elements were present, the age at death of adult individuals was estimated using the pubic symphysis scoring system according to Brooks and Suchey (1990), the auricular surface of the ilium according to Lovejoy et al. (1985), the sternal articular surfaces of the clavicle (Szilvássy 1978). Furthermore, the closure of cranial sutures (Meindl and Lovejoy 1985) and mandibular and maxillary attrition were analyzed according to Lovejoy (1985).

Sex/gender determination

For biological sex determination, pelvic bones were primarily used following the Phenice method (Phenice 1969) and the classification scheme according to Walker (2005). In addition, skull morphology was used for sex determination, according to Walker in Buikstra and Ubelaker (1994). Furthermore, biological sex was determined using 28 selected characteristics of the skull and mandible, the pelvis, and the robustness of the humerus and the femur according to the catalog of characteristics by Acsádi and Nemeskéri (1970) and Ferembach et al. (1979) The degree of expression of these 28 characteristics was rated on a scale from -2 (hyperfeminine) to +2 (hypermasculine). In individuals under the age of 18, the sex was only determined if the sex-determining characteristics mentioned above were already clearly developed.

Besides biological sex, gender was estimated based on grave goods. Weapons and knives were associated with the male gender, while needles, specific jewelry, and mirrors were associated with a female identity. Since biological sex determination and archeological gender estimation differed only in one individual, in the present study, biological sex determination was used. This match of biological sex determination and gender-typical grave goods can be cautiously interpreted as evidence for a binary sex/gender system. Furthermore, among 10 adult individuals, an aDNA analysis was performed to verify the molecular sex of the individuals. The morphological sex determination and the molecular sex determination corresponded to 100%.

To quantify the ratio of male and female-determined individuals, the sex ratio was calculated. The sex ratio is commonly defined as the number of men to every 100 women (Hobbs 2004).

sex ratio= 100*n men/n women

The sex ratio can be expressed in units (1.02) or hundreds (102) to demonstrate the number of men per 100 women. In the present study, we used units.

Statistical and spatial analysis

Statistical analyses were carried out using SPSS Version 24. After computing descriptive statistics, differences in the sex proportion between the burial areas, the age groups, and the chronological periods were calculated by means of Chi squares. A *p*-value below 0.05 was considered significant.

The entire dataset has spatial components. Therefore, a geographic information system (GIS) plays an essential role in the research design, as it can ideally accommodate our sites' complex relational and diachronically changing contexts. In addition to digital cartography, the GIS serves primarily as a central database and and analysis tool for all data, including open government data (OGD). Consequently, the study relies on the records of archaeological contexts, the recorded human remains, and their bioarchaeological analysis results for the digital and contextual archaeothantological analysis of (bio)archaeological data in a comprehensive geodatabase. For this purpose, an integrated digital approach was applied, using both proprietary and free and open source software (FOSS) for information management and spatial data analysis (Hagmann 2020). The GIS software was QGIS (latest 3.30 's-Hertogenbosch) installed on a Windows 11 Pro device (as SPSS). As a coordinate reference system, we used the Austrian National Survey Coordinate System for the central part of Austria, EPSG:31255 MGI / Austria GK Central (https://epsg.io/31255). In the first step, all digital and analog excavation plans were imported into the GIS or (in the case of analog data) retro-digitized and then geo-referenced. The digital general archaeological map of the city of Wels provided the overall basis for spatial analysis (Greisinger et al. 2020). Then, in step 2, all burial pits were mapped in the GIS to obtain a complete dataset of all burials in Ovilava's eastern cemetery, also integrating data from the digital general map. In step 3, significant qualitative information was selectively extracted from the excavation documentation records and mapped onto the newly generated geospatial dataset utilizing standardized attributes. For this purpose, cultural data was collected in structured tables in Excel/Microsoft 365 (XLSX file format), and a sophisticated folder structure was used for sustainable data management. All data was securely stored in a cloud (Google Drive). Spatial vector data stored in ESRI shapefiles (SHP and auxiliary files) provided all quantitative technical information about the area of interest. In step 4, the qualitative archaeological dataset was merged with the systematic radiocarbon dating data and loaded into the FOSS GIS software as non-spatial tables in CSV file format. Further, the quantitative and qualitative bioarchaeological attributes were merged with those of the spatial cemetery dataset to obtain the final dataset. For this purpose, the cultural-qualitative tables were merged with the spatial-quantitative information, followed by validation activities regarding quality control. The results were used for map-based cartographic visualization and plotted in TIFF file format; by that, we applied a specific form of integrated 'deep-mapping'-approach for visualizing archaeological data, providing a richer, more nuanced, multilayered, and multidimensional understanding of the burial ground per each map (Hagmann 2020). (Hagmann 2020, 2021; Kirchengast and Hagmann 2022).

Results

Spatial analysis of the burial ground

The four defined burial areas differed highly significantly ($p<0.003$) in temporal patterns. While most of the burials in the Kanalgrabung area dated to early medieval times (63.6%), the Alois-Auer-Straße (100%) and Dr.-Groß-/Dr.-Schauer-Straße (50%) areas were predominantly used in Roman Imperial times. More than 70% of the Fronius area individuals dated in the Roman era, while the area of the Pendlerparkplatz was used also mostly used in Imperial Roman times (62.2%) but also the Early Roman (20%) and Early Medieval time (17.8%) (see Table 1 and Figure 3).

Table 1. Description of the individual burial areas at the Gräberfeld Ost

area	Period			Burial type		
					inhumations	
	ER	IR	EM	cremations	graves	individuals
	n	n	n	n	n	n
Pendlerparkplatz	9	28	8	116	61	77
Fronius	8	24	1	186	50	51
Kanalgrabung Dr.-Groß-Straße	1	3	7	20	15	17
Alois-Auer-Straße	0	2	0	0	5	5
Dr.-Groß-/Dr.-Schauer-Straße	1	1	0	6	3	3
total	19	58	16	328	134	153

Legend: ER = Early Roman, IR = Imperial Roman, EM =Early Medieval

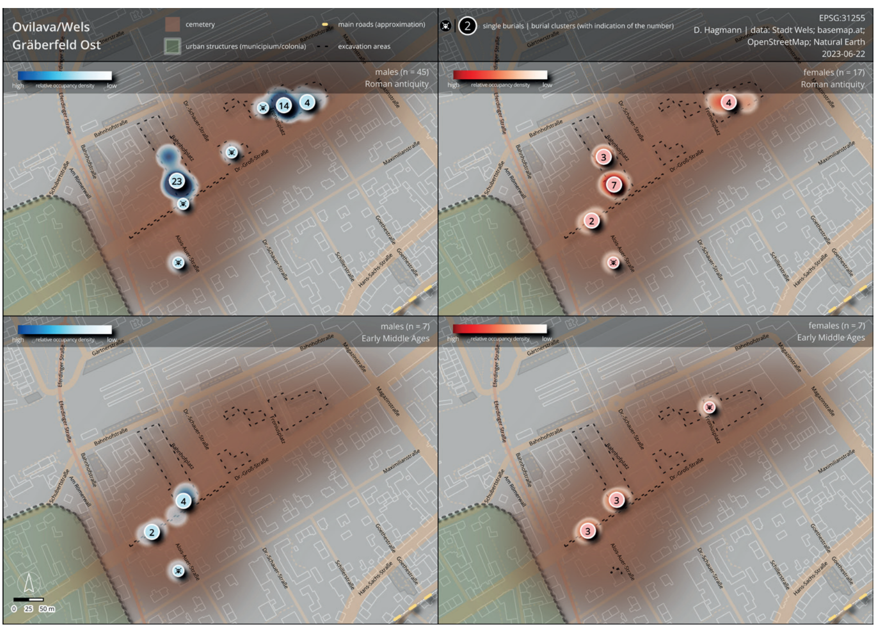

Fig. 3. Spatio-temporal occupancy patterns of the defined excavation areas, segregated by male and female inhumation burials, in relation to Roman Antiquity and the Early Middle Ages

Age distribution

More than 30% of the individuals in the inhumation sample were subadults, with a particularly high percentage of regularly buried neonates (11.7%). 16.5% of the subadult individuals were classified as infans I or infans II, indicating an age at death between 2 months and 13 years. The majority of the individuals buried at the Gräberfeld Ost areal died between the ages of 20 and 40 years (42.6%). 22% died between 40 and 60 years, and only 2 % were older than 60 years (Figure 4).

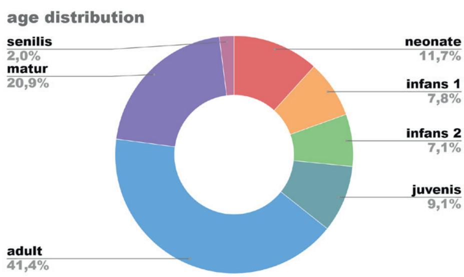

Fig. 4. Age distribution of the whole sample

Sex ratio

The biological sex distribution of the whole sample is presented in Figure 5. Considering the individuals classified as juvenile or adult only, the number of male-classified or likely male-classified individuals significantly surpassed the number of female-classified or likely female-classified ones (see Figure 5). The sex ratio of the whole sample is 200. This observation means that the proportion of men in this burial ground is twice as high as that of women.

However, when the individual cemetery areas, age groups, and chronology are considered separately, it becomes apparent that this marked female deficit varies temporally and spatially. As shown in Table 2, the respective cemetery areas differ concerning the sex ratio. A female deficit can be observed for the Fronius, Pendlerparkplatz, Alois-Auer-Straße, and Dr.-Groß-/Dr.-Schauer-Straße areas. No female-classified skeleton was excavated in the Dr.-Groß-/Dr.-Schauer-Straße-area; however, this area yielded only 3 adult individuals. Considering the larger burial areas, the Fronius area showed the highest female deficit (sex ratio = 287.5).

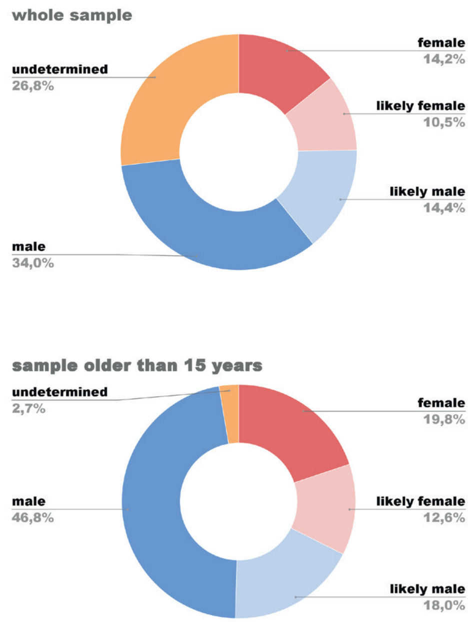

Fig. 5. Sex distribution of the whole sample (n=153) and of the individuals older than 15 years (n=110)

In the Pendlerparkplatz area, almost twice as many males are buried as females. Only the Kanalgrabung area revealed more female than male skeletons. When comparing the sex ratio across different age groups - juvenile (15 to 19 years), adult (20 to 40 years), and mature (>40 years) - a noticeable surplus of female-classified skeletons is observed in the youngest age group. However, a female deficit emerges in the adult age group. In the highest age group, male-classified skeletons outnumber female ones by almost four to one (see Table 2).

In a further step, the time periods were compared. Here, the significantly highest surplus of men was observed in the Early Roman Period (1st cent. AD). Furthermore, a significant female deficit could be observed in the Roman Imperial Period (2nd and 3rd cent. AD) – more than twice as many male-classified skeletons could be found. Only in the Early Middle Ages (7th and 8th cent. AD) the sex ratio was almost balanced, although a slight surplus of males could still be observed (see Table 2).

Table 2. Sex ratio (100*M/F) according to burial area, age group, and period

	F/F?		M/M?		UND		Total	Sex ratio
	n	%	n	%	n	%	n	(men to every 100 women)
Areal								
Kanalgrabung	7	53.8%	6	46.2%	0	0.0%	13	85.7
Pendlerparkplatz	20	32.8%	38	62.3%	3	4.9%	61	190.0
Fronius	8	25.8%	23	74.2%	0	0.0%	31	287.5
Alois-Auer-Straße	1	33.3%	2	66.6%	0	0.0%	3	200.0
Dr.-Groß-/Dr.-Schauer-Straße	0	0.0%	3	100.0%	0	0.0%	3	.
Chi-square			12.2	*p*=0.429				
Total burial ground	36	32.4%	72	64.9%	3	2.7%	111	200.0
Age groups								
Juveniles 15 to 19 yrs	8	57.1%	5	35.7%	1	7.1%	14	62.5
Adults 20 to 40 yrs	18	37.5%	29	60.4%	1	2.1%	48	161.1
Mature > 40 yrs	10	20.4%	37	75.5%	1	4.1%	48	370.0
Chi.square			5.26	*p*=0.218				
total	36		71		3		110	
Period								
Early Roman	4	22.2%	14	77.8%	0	0.0%	18	375.0
Imperial Roman	15	30.0%	35	70.0%	0	0.0%	50	233.3
Early Middle Ages	7	46.7%	8	53.3%	0	0.0%	15	114.3
Chi-square			14.4	p=0.026				
total	26	31.3%	57	68.7%	0	0.0%	83	219.2

legend: F/F? = female and likely female, M/M? = male and likely male; UND =undetermined

In the last step, the sex ratio of the eastern Ovilava burial ground was compared to those of other Roman times samples from Austria, Germany, Hungary, Serbia, and North Macedonia. As presented in Table 3 and Figure 6, a female deficit occurred in many samples. Only the Austrian samples from Saladorf (Roman rural *villa* population) and Petronell-Carnuntum (Roman urban population next to the legionary fortress), as well as the Hungarian sample from Pécs (Roman urban population of Sopianae), revealed a female surplus. All other samples showed a marked female deficit (see Table 3 and Figure 6).

Table 3. Sex ratio of Ovilava and Roman times samples (inhumations only) around the Danube River Basin (adapted from Schweder & Winkler 2004, 73 tab. 39 and supplemented by the authors)

Roman samples		F/F?	M/M?	Sex ratio (men to every 100 women)
Austria				
Saladorf[1]		12	9	75.0
Carnuntum[2]		24	19	79.2
Carnuntum	Mühläcker[3]	7	7	100.0
Lentia/Linz	Tiefer Graben[4]	16	16	100.0
Mannersdorf a. Leithagebirge[1]		10	12	120.0
Halbturn[5]		24	30	125.0
Brunn a. Gebirge[6]		4	5	125.0
Vindobona/Vienna-Albertina	Albertina[7]	20	26	130.0
Comagena/Tulln a. d. Donau[8]		14	19	135.7
Arrianis (?)/Klosterneuburg[9]		10	18	180.0
Lauriacum/Enns	Espelmayrfeld[10]	34	65	191.2
Ovilava/Wels		36	72	200.0
Pottenbrunn[11]		15	36	240.0
Oggau a. Neusiedlersee[12]		4	10	250.0
Lauriacum/Enns	Steinpaß[13]	54	143	264.8
	Ziegelfeld[14]	33	107	324.2
Germany				
Neuburg a. D.	Zone 1[15]	11	32	290.9
Neuburg a. D.	Zone 2[15]	9	27	300.0
Neuburg a. D.	Zone 3[15]	3	10	333.3
Stettfeld[16]		4	18	450.0
Switzerland				
Bonaduz[17]		190	269	141.6
Hungary				
Pécs[18]		52	37	71.2
Tokod[19]		50	56	112.0
North Macedonia				
Deboj-Ohrid[20]		21	45	214.3
Serbia				
Viminacium	Pećine [20]	65	144	221.5
Viminacium	Više Grobalja[20]	11	38	345.5

Legend: 1 Pail 2009; 2 Schweder & Winkler 2004; 3 Harrer 2014; 4 Wiltschke & Teschler-Nicola 1991; 5 Berner 2014; 6 Szilvassy 1976; 7 Marschler 2013; 8 Lebzeltner et al. 1935; 9 Neugebauer et al. 1987; 10 Kloiber 1962; 11 Hölbling 2008; 12 Ehgartner 1947; 13 Pichelmayer 1972; 14 Kloiber 1957; 15 Ziegelmayer 1979; 16 Wahl 1988; 16 Knötzele 2018; 17 Brunner 1972; 18 Ery 1973, 19 Ery 1981, 20 Mikic 1984

Fig. 6. Geospatial location of Ovilava and Roman times samples (inhumations only) regarding sex ratio across the Danube River Basin

Discussion

In the present study, we analyzed the tertiary sex ratio of the inhumations of the eastern cemetery of Ovilava only. Before we start with a detailed discussion of the results, we must mention the limitations of the present study. First, it should be noted that the entire cemetery of Ovilava could not be excavated, as it is now located under the densely built-up urban area of the city of Wels in Upper Austria. It is, therefore, only possible to analyze individual areas uncovered randomly during rescue excavations. Therefore, the number of skeletons included in the analysis, 111, is relatively small. The analysis, however, included only inhumations, and the more than 328 cremations have yet to be analyzed. Although there is no hint from the literature that burial practices (inhumations versus cremations) varied significantly between men and women in the time periods considered in this study, the fact that cremations are not included in the analysis represents a major limitation of this study (*infra*).

Furthermore, regarding the general spatial location of the study area, it is unclear whether the road, which continued eastward from the inner-city *decumanus*, was the main exit road or just another suburban feeder parallel to the principal axis. Therefore, it remains to be considered whether the present reconstructed picture corresponds at all to the full extension. It can also be assumed that the actual core areas of the cemetery are still archaeologically unrecorded and lay further southeast around the *decumanus maximus* (east-west main road) and a main drop-off road adjoining it, which so far could not be further identified archaeologically. Accordingly, it is also possible that the currently reconstructed area of the Gräberfeld Ost represents only a peripheral section of the outermost edge of a much larger cemetery. Further, it must be noted that radiocarbon dating has shown that inhumations occurred in the cemetery during the entire Roman Antiquity, even if during the Early Roman Period (seemingly in favor of cremations), the inhumations occurred only sporadically. Consequently, this result does not change the generally accepted assumption that inhumations only began to appear more frequently c. at the end of the 2nd cent. and largely replaced cremations, but it must be emphasized that inhumations should not be placed due to this premise in the later Roman periods from the outset (*infra*).

The tertiary sex ratio represents a crucial demographic parameter that quantifies a population's numerical ratio of male to female individuals. The analysis of the sex ratio of historical populations poses a particular challenge, as the numerical sex ratio can only be determined utilizing written records, such as gravestone inscriptions, or the bioarchaeological analysis of burial grounds. Since sex-typical features on the skeleton that allow sex diagnosis do not manifest until puberty and adolescence, in most cases, osteological sex diagnosis can only be made from the juvenile phase onwards. Furthermore, poor preservation status reduces the possibility of a valid sex diagnosis.

The good state of preservation of the skeletons of the eastern Ovilava cemetery allowed a morphological sex estimation for the 108 older juvenile and adult individuals and, thus, hypothesis testing. Hypothesis 1 could be verified. Of the 108 individuals at least 15 years old, only 36 could be classified as likely female or female, while 72 skeletal individuals were determined to be likely male or male. Only for three skeletons, sex estimation was impossible due to the poor state of preservation. Therefore, a sex ratio of 200.0 among the inhumations could be determined for the sample older than 15 years, which means twice as many males as females among the inhumations at the Ovilava cemetery. This enormous male surplus corresponds to those of numerous other historical samples, especially from the Roman period:

As shown in Table 3, a sex ratio of 200.0, as in Ovilava, does not represent the most pronounced male surplus or female deficit. Except for the samples from Saladorf (Pail 2009) and Petronell-Carnuntum (Schweder and Winkler 2004), the majority of Roman samples from Austria (Lebzelter 1935; Ehgartner 1947; Kloiber 1957, 1962; Pichlmayer 1972; Neugebauer-Maresch and Neugebauer 1987; Berner 2014; Marschler 2013), Germany (Ziegelmayer 1979; Wahl 1988), and Switzerland (Brunner 1972), but also in the former Yugoslavia (Mikic 1984) had a sex ratio above 100.0 and thus a female deficit. Only for Lentia (Wiltschke and Teschler-Nicola 1991) and Carnuntum Mühläcker (Harrer 2014), a sex ratio of 100, indicating as many females as males, was reported. Consequently, a female deficit seems a not unusual demographic pattern among inhumations in the Roman *limes* provinces.

In most older studies, the female deficit of historical populations was assumed to be an unquestioned fact; in new studies, methodological errors are often discussed as explanatory approaches (Cintas-Peña and Herrero-Corral 2020). Furthermore, a poor state of preservation of the female skeletons, which may lead to misidentifications, was postulated (White and Folkens 2005). Also, the possibility is discussed that gracile female skeletons would not have been preserved at all, and thus an unnatural sex proportion is justified. The present sample from Ovilava does not support this interpretation. The majority of the skeletons were in an excellent state of preservation, which allowed a morphological sex diagnosis. In addition, 10 aDNA analyses of skeletons from Ovilava are available to date, which had been analyzed as part of another study. All chromosomal sex diagnoses corresponded to the morphological sex estimations. Furthermore, the morphological sex estimation was compared with the grave goods. Here too, a correspondence between the biological sex and the gender associated with the material remains was found. Therefore, there is no indication of a methodological misdiagnosis.

Considering that only the inhumations, and not the cremations, have been included in the analysis, we must state that a complete demographic analysis would only be possible by including all burials, including both inhumations and cremations. Consequently, an obvious explanation for the female deficit would be that Roman women in Ovilava were cremated more often than their male counterparts. However, no evidence from historical sources suggests that burial practices differed between sexes in a standardized manner nor that sex-specific, separate burial grounds were systematically used in the Roman Empire (Barber & Bowsher 2009; Gonçalves et al. 2010). Already in Ovilava, the well-known grave portraits of a married couple, dating back to the Roman Imperial Period (Antoninian/2nd cent. AD) and documented since the 16th cent., are prominently embedded in a house facade on the main square. They depict a married couple in the form of half-figures. In their original context, it is assumed that they were part of a more elaborate funerary monument, likely placed at a family burial site, indicating that individuals of all genders were interred there (http://lupa.at/593).

Moreover, in the case of the Pendlerparkplatz area, inhumation and cremation burials do not intermingle in the overall picture, except for very few possible exceptions (Figure 2). The burials, instead, tend to be distinctly separate from each other. For most cases, this observation can be understood as no mixing of cremated and physically buried individuals in the same plot. Instead, it seems that both males and females were either both cremated or physically buried. The general and precise situation regarding burial customs and rites in the Roman Empire over time remains generally unclear due to a lack of comprehensive data evaluation. Overall, one should always consider regional and zeitgeist-influenced specifics, as the explicit, 'typically Roman burial custom' in this particular form did not exist (Pearce 2011; Prowse 2014). While the 'Gräberfeld Ost' exhibits a bi-ritual nature, it is currently difficult to foresee which patterns allow us to distinguish between cremation and inhumation burials regarding timing and funerary practices for each individual. The traditional division between early cremations until approximately the 2nd cent. AD and later body burials from approximately the 2nd cent. AD onwards (Darby Nock 1932; Toynbee 1996) does not appear to be entirely applicable in Ovilava, as supported by radiocarbon dates (*supra*). This situation necessitates exploring other explanatory approaches (Pearce 2016).

Another interpretation of the female deficit may be the higher mortality rate of female newborns and children, which could have led to a deficit of adult females. Due to the low validity of morphological sex diagnoses on the subadult, especially prepubertal skeleton, this interpretation cannot be confirmed but also not refuted. Infant mortality was undoubtedly high in Roman times, but whether more girls died cannot be said. According to the male-disadvantage hypothesis, however, the male sex would be the more vulnerable and affected by a higher risk of morbidity and mortality (Naeye 1971; Kirchengast and Hartmann 2009; Pongou 2013). The male disadvantage hypothesis contrasted with an assumed lower social value of female children in the very patriarchal Roman society, which may have resulted in higher death rates of female children, whether by active infanticide or by deliberate neglect leading to higher mortality of girls (Boswell 1988; Harris 1982, 1994). Several other studies, however, found no evidence of increased female infanticide rates (Engels 1980; Haentjens 2000); the possibility of female infanticide, however, cannot be ruled out (Scheidl 2010). In the present sample, higher mortality of females in the subadult age group was thus evident during adolescence. If increased mortality was evident at prepubertal age, too, this could lead to a shift in the sex ratio. On the other hand, skeletal evidence from Roman time cemeteries clearly showed that the majority of children, independent of their sex, suffered from severe stress phases caused by malnutrition or diseases resulting in visible skeletal stress makers such as Cribra orbitalia, enamel hypoplasias or even stunting (Pilkington 2013). However, high mortality rates among newborns and children can be assumed for both sexes. An indication of a female deficit at the marriageable age may be the marriage legislation under Augustus in the 1st cent. AD. Here, non-senatorial freeborn men are possibly encouraged to marry freed women, suggesting a shortage of women, at least among the Roman upper classes (McGinn 2004). However, this lack of women could also be due to the excess mortality of women of reproductive age. Pregnancy and childbirth represent dramatic mortality risks in the recent past and even today (Alkema et al. 2016; Hirshberg and Srinivas 2017).

Nonetheless, since the bioarchaeological evidence for deaths causally related to reproduction is extremely rare (Stone 2016), the interpretation here can only be indirect. The higher number of female skeletons in the juvenile age between 15 and 19 years could be interpreted concerning the increased mortality of females in relation to reproduction. Also, the female deficit is lower in the age class between 20 and 40 years, consequently in the reproductive phase than in the post-reproductive phase. This observation also proves the verification of the second hypothesis. In the present study, it was shown that the sex ratio varies both as a function of age classes and between periods. Comparing the Early Period, i.e., the 1st cent. AD, with the Roman Imperial Period and the Early Middle Ages shows a reduction of the female deficit over time. The most prominent female deficit is found in the Early Period, which coincides with the land occupation and foundation of Ovilava. In this phase, men outnumber women by a factor of three. In the Imperial Period, the female deficit was reduced, and in the Early Middle Ages, the sex ratio was almost balanced, corresponding to other early medieval samples such as the Avar burial ground of Csokorgasse, Vienna (Grossschmidt 1990).

Although the proportion of early medieval skeletons in the cemetery of Ovilava is very low, a clear trend towards a balanced sex ratio can be recognized. This condition could be attributed to the increased societal value of the female sex on one hand, and to ecological changes on the other. After the RCO between 250 and 400 AD, temperatures dropped, and a more unstable climatic situation set in in the area of the Roman Empire. Studies of recent populations show that unstable climatic conditions lead to increased maternal stress and may lead to an increase in the sex ratio, as male fetuses are more vulnerable to stress, increasing miscarriages and stillbirths of male fetuses, while female fetuses prove to be more resistant to stress (Fukuda et al. 2014). The end of the RCO in the 3rd cent. AD would be possible as an explanatory model here, as a decline in average temperatures in southern and central Europe may have negatively affected the ratio of male to female neonates, thus reducing the female deficit. In the present study, there is a reduction in the female deficit with increasing timing. In the other studies, which cite an apparent female deficit in Roman-period samples, this aspect cannot be verified since no dating of the individual individuals of the sample was carried out. However, mostly only a chronological classification was made based on material legacies. Stress factors such as plagues, economic changes, and increasing insecurity at the *limes* borders (McCormick et al. 2012; Pilkington 2013; Il and Joksimovic 2015; Wippert et al. 2017; Zuckermann and Crandall 2019) could also have increased the stress for expectant mothers and infants and thus led to a reduction of more vulnerable male fetuses and consequently a reduction of the female deficit.

Another explanatory model for the female deficit at sites of the *limes* region could be based on the legionary camps of the region, where females, despite their regular on-site presence, were a minority *per se*. However, such an explanation cannot be assumed for the present study since Ovilava did not represent a legionary camp but clearly a 'rural-civilian' town in the hinterland. For example, a similar picture can also be seen for Pottenbrunn (Hölbling 2008), which represents a distinct rural civilian settlement (probably a *vicus*), having an even lower sex ratio than Ovilava.

The present study tested another explanatory option as part of a spatio-temporal analysis. The second hypothesis predicted a variation in the sex ratio between the individual burial areas and could be verified. In a spatial-temporal analysis, the individual cemetery areas were compared, and it was shown that, on the one hand, the cemetery areas were mainly used at different times, so the Early Medieval skeletons were mainly found in the area of canal excavation and, therefore a surplus of women in the Kanalgrabung area as well as a balanced sex ratio in the Early Middle Ages. Both of these findings are associated. The idea that males and females have been buried in different sections of the burial ground could be rejected because male and female individuals were found in all burial ground areas. Finally, such a model would have contradicted specifically the general evidence in most other places regarding Roman burial customs (Scholz 2022).

An extraordinary male surplus could be found for Alois-Auer-Straße and Dr.-Groß-/Dr.-Schauer-Straße, but each area contains only 3 individuals in total and is therefore of little significance. However, the area of Fronius, which is of particular interest and is located furthest from the town of Ovilava with a sex ratio of 0.35, exhibited a much higher male excess compared to the Pendlerparkplatz area, which had a sex ratio of 0.53. The Fronius area dates primarily to the Early and Roman Imperial Period and is characterized by a high proportion of skeletons with severe wear on the spine and joints (Kirchengast et al. 2023). Although these severe stress features are observable in male and female classified skeletons, this finding argues for a hard-working population composed predominantly of males. Indicators of increased somatic stress in the *limes* hinterland could also be demonstrated for skeletal series from Croatia. Here, too, men show particularly frequently load-induced pathological changes of the spine (Šlaus et al. 2003). The fact that men affected by particular physical stress were mainly buried in a specific decentralized area of the cemetery (Fronius area) allows the following cautious interpretation:

The urban center of Ovilava was an attraction point for migrant workers or labor migrants from nearer and farther regions (de Ligt and Tacoma 2016; Lo Cascio and Tacoma 2016). Nevertheless, a planned project with isotopic investigations and aDNA analyses should provide clarity here in the future. We interpret the results of the spatial analysis as indicating an organized funerary system, presumably in the form of private burial associations (*collegia funeraticia*) and possibly even municipal magistrates as proxies, to provide burial in distinct, remote, and therefore possibly favorable areas of the burial ground. They may have ensured that, after a fatal accident or a lonely death on the street, the human remains of workers, the poor, and even outcasts, as well as other accidental deaths without relatives, were recovered and given a dignified burial. This interpretation, also considering the hygienic aspects and the overriding social attitudes regarding death pollution, is further supported by the fact that we do not observe any of the much-quoted but hardly documented mass graves for the poor (*puticuli*) but discrete individual burials in the "potter's fields" of Ovilava (Bodel 2000; Emmerson 2020; Graham 2006a, 2006b; Perry 2017; Sano 2012; Verboven and Laes 2016). The possible presence of a larger group of poor, predominantly male migrant workers or labor migrants, may have contributed to the observed female deficit or male surplus in the Ovilava inhumations (Le Guennec and Stevens 2020; Séguy 2019; van Driel-Murray 2008; Verhagen 2019) and may also be supported by contemporaneous observations: The most pronounced male surplus in the world is currently found in Qatar with a sex ratio of 287.0, which is comparable to the findings of the Fronius area of Ovilava. This exorbitantly high male surplus in Qatar is explained by the high number of impoverished male migrant workers and labor migrants living in Qatar and not by a female deficit of Qatar's autochthonous population. Similar findings could be described for the Arab Emirates, Saudi Arabia, and Oman (UNO 2022).

Conclusion

Since only inhumations and no cremations have been included in the analysis, we must clearly state that this is a pilot study only. The analysis of the sex ratio of the civilian Roman city of Ovilava in the *limes* hinterland reveals an extremely unbalanced sex ratio in favor of the biological male sex. This situation was especially true of the Early Roman and Imperial Roman periods, but not for early medieval times. A female surplus was only found for the youngest age group aged between 15 and 19 years. Similar findings are reported for many other Roman times cemeteries in the *limes* hinterland. In the present sample, the spatial analysis of the sex ratio revealed an extremely high male surplus for the so-called Fronius part of the cemetery, an area that a physically hard-working population may have occupied. Therefore, the spatial analysis may allow a socioeconomic interpretation of the unbalanced sex ratio of the cemetery of Ovilava. Further studies, including aDNA and stable isotopes, are necessary to clarify the origin of the individuals buried at the Ovilava burial ground.

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Conflict of interest

The authors declare no competing interests.

Authors' contribution

D.H. designed the study, carried out the spatial analysis, and wrote the manuscript, B.A. did the osteological analysis, M.G. excavated the burial ground and provided information regarding material culture, R.M. excavated the burial ground and provided information regarding grave goods and history, S.K. designed the study, did the osteological analysis and wrote the manuscript

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