

SLAJ

Volume: 5; Issue: II; 2021

*The Official Publication
of the
Anatomical Society of Sri Lanka*



e-ISSN 2550-2832



Sri Lanka Anatomy Journal

Journal of the Anatomical Society of Sri Lanka

Volume 5: Issue II: 2021

The Sri Lanka Anatomy Journal is a peer reviewed journal published two times a year by the Anatomical Society of Sri Lanka.

Editorial Office

Department of Anatomy,
Faculty of Medical Sciences,
University of Sri Jayewardenepura,
Sri Lanka.

Correspondence regarding editorial matters, articles, reviews and news items regarding the membership should be addressed to the Editor, SLAJ, Dr. Sithara Dissanayake, Department of Anatomy, Faculty of Medical Sciences, University of Sri Jayewardenepura, Sri Lanka.

E-mail -

srilankaanatomyjournal@gmail.com

CC BY 4.0

This is an Open Access article distributed under the terms of the Creative Commons Attribution 4.0 license (unless stated otherwise) which permits unrestricted use, distribution and reproduction in any medium, provided the original work is properly cited. Copyright is retained by the author(s).

e-ISSN 2550-2832

Editorial Board

Editor

Dr. Sithara Dissanayake

Assistant Editor

Dr. Sajith Edirisinghe

Editorial Board

Professor S.G. Yasawardene

Professor B.G. Nanayakkara

Professor Isurani Ilayperuma

Professor D.J. Anthony

Professor Madhuwanthi Dissanayake

Professor Ajith Malalasekara

Professor Harsha Dissanayake

Dr. Jayampathi Dissanayake

Dr. Varuni Tennekoon

International Reviewer

Professor Simon H. Parson FAS

Regius Professor of Anatomy,

Anatomy, Suttie Centre,

University of Aberdeen,

Foresterhill, Aberdeen, AB25 2ZD

Advisors

Emeritus Professor Rohan W. Jayasekera

Emeritus Professor L.S.S. Salgado

Managerial Assistant

Mr. Chamil Kumara

CONTENT

Editorial

1. Promoting Communication Skills during Basic Science Teaching in the Medical Curriculum
Madhuwanthi Dissanayake, Professor & Consultant Eye Surgeon, Department of Anatomy, Genetics & Medical Informatics, Faculty of Medicine, University of Colombo

Review Article

2. Involvement of Parathyroid glands in COVID-19
Niranjana R

Original Research

3. An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka
KM Chandimal, K Sirak, EAST Edirisinghe, G Adikari, D Reich and SG Yasawardene
4. Morphometric Analysis of Dry Human Mandibles of Sri Lankans
KM Chandimal, Amila Wickramrathna, PD Wimalasinghe, DM Ranasinghe, SG Yasawardene

Case Reports

5. Isolated giant mesenteric vein and anomalous inferior mesenteric vein insertion in the presence of cirrhosis - A cadaveric case report
Jayarathna MJS, Udayakumara S, Dhanushkar R, Weerasinghe S, Amaratunga HA

Instructions to Authors

EDITORIAL

Promoting Communication Skills during Basic Science Teaching in the Medical Curriculum

Outcome-based medical education (OBME) is a widely accepted approach in medical education. It has been implemented in various forms in many medical schools and residency programs around the world. Practiced in many countries, outcome - based medical education focuses on several key areas, including:

1. Patient care: high-quality, patient-centered care, consistent with current standards of practice.
2. Medical knowledge: thorough understanding of the medical knowledge required to provide appropriate care to patients.
3. Clinical skills: demonstrate proficiency in the clinical skills required to diagnose and treat patients.
4. Interpersonal and communication skills: effectively communicate with patients, families, and other healthcare providers.
5. Professionalism: demonstrate the values and behaviors of a responsible and ethical healthcare professional.
6. Systems-based practice: knowledge and skills to effectively navigate and improve the healthcare system in which they practice.
7. Continuous quality improvement: ability to critically evaluate their own performance and make necessary changes for improvement.

8. Inter-professional collaboration: ability to work effectively with other healthcare professionals from different disciplines.

Of the main outcomes, medical knowledge and clinical skills have traditionally been the main focus of the medical curriculum in many medical schools. However effective communication skills are essential for success in the medical field, and its importance should be emphasized throughout the entire medical education process.

Contrary to the common belief, there can be many opportunities for the communication skills to be promoted and developed during the early years in the medical school.

Developing communication skills, including reading, writing, speaking and listening in formal or informal settings will empower the undergraduate giving confidence and self-satisfaction from an early point in his or her career.

There are numerous methods to promote communication skills in the first year of medical school during the teaching of basic sciences. A conducive environment showing sensitivity to cultural, social, and linguistic diversity will promote and encourage the new entrant to engage in the activities in promoting communication skills. These activities are best done incorporated with the real-world scenarios in collaboration with the basic sciences teaching and learning activities. Small group activities / discussions and problem-

based learning sessions are ideal settings. This can include providing opportunities for students to practice communication skills in their day to day practice in the academic settings as well as in early clinical exposure settings, interacting with patients and other healthcare professionals.

Active listening is an essential component of effective communication, so it is important to encourage students to practice active listening and to give and receive feedback on their communication skills as well.

Although not emphasized enough, dissections and practical sessions serve as a good setting as the communication is driven by the learning task during the group activity.

Student seminars where students make preparations and presentations on their own under the guidance of an academic around a clinical scenario is another golden opportunity for promoting communication skills.

Providing opportunities for extra-curricular activities such as quizzes, debates, public speaking, exhibitions for the public, drama and theater groups and community projects can help students develop their communication skills in a more relaxed and low-pressure setting. With the advent of computer based and IT related tools, students are far head in utilizing many applications which help them to enhance their communication skills.

Focusing on assessments, both formative and summative forms play an important role. Especially while practicing for these, students

will further enhance their communication skills. Viva voce examinations conducted in a structured manner and in a friendly setting will be an excellent way of improving one to one communication capabilities.

Teachers in basic sciences should consider communication skills as an important program learning outcome and should not only grab each opportunity to promote communication skills but also document and incorporate them in the written curriculum for the perusal of reviewers and accreditors.

Prof. Madhuwanthi Dissanayake,
Professor & Consultant Eye Surgeon,
Department of Anatomy, Genetics & Medical Informatics,
Faculty of Medicine,
University of Colombo.

CC BY 4.0

This is an Open Access article distributed under the terms of the Creative Commons Attribution 4.0 license (unless stated otherwise) which permits unrestricted use, distribution and reproduction in any medium, provided the original work is properly cited. Copyright is retained by the author(s).



REVIEW ARTICLE

Involvement of Parathyroid gland in COVID-19

Nirajan R¹

¹Department of Anatomy, Faculty of Medicine, University of Jaffna, Sri Lanka

Abstract

Introduction: Parathyroid glands are yellow brown, small and soft. Typically four parathyroids are present in posterior surface of thyroid. Parathyroid's numeral, size and site is inconstant. Corona virus disease-19(COVID-19) is a major problem all over world and aim of this study is to discuss possible involvement of parathyroid in COVID-19.

Materials and methods: The previous studies were used for this review study.

Results and discussions: The COVID-19 mainly causes pulmonary injury and respiratory failure. Virus was chiefly detected in respiratory pathway, gastrointestinal tract, sweat gland, pituitary, pancreas, adrenal, renal, parathyroid, liver and cerebrum. Nearly 65% of COVID-19 patients had hypocalcaemia. Advanced age and hypocalcaemia were bad prognostic factors for COVID-19 patients. Hypocalcaemia might be due to vitamin-D deficit, decreased bowel absorption of calcium, diminished secretion or response to parathyroid hormone (PTH). The PTH rises serum calcium by discharge calcium by osteoclastic activity, calcium reabsorption in nephron. Vitamin-D encourage reabsorption of dietary calcium. Thus inadequate Vitamin-D excites parathyroid to discharge more PTH.

Normal and abnormal parathyroids are identified by their size, form, consistency and

histological judgments. There was association between size of parathyroid and PTH level. Glands of black populations were longer than white groups. Larger parathyroid secretes more PTH and might has greater number of chief cells. Microscopically quantity of fatty tissues and oxyphil cells rise in elders. Both transitional and oxyphil cells secretes PTH to extensive parathyroid stimulation.

Conclusion: Size of all parathyroids, patient's age, oxyphil cells amount, ethnicity, amount of PTH and Vitamin-D determines the serum calcium level in COVID-19.

Keywords: Parathyroid involvement, COVID-19, Vitamin-D therapy, Parathyroid hormone.

Introduction

Parathyroid glands are smaller endocrine structures and typically four glands are present in every individual with two on each side but the numeral, size and site of parathyroid glands is inconstant (1). The parathyroid gland commonly lies near to the posterior surface of thyroid gland underneath a thin film of fascia, encircled by fat globules. Parathyroid gland itself has a thin capsule. The parathyroid is a minor yellow-brown structure within the range of 35 -40 mg weight and 5×3×1 mm in dimension (2). Normal parathyroid glands are

light and pliable in consistency. This helps in distinguishing it from the other nearby anatomical structures (3). Lymph nodes might be firmer than the parathyroid glands.

The severe acute respiratory syndrome coronavirus (SARS- COV) has diseased millions of people and it creates a major community problem, cost-effective and health complications all over the world (4). It was documented about the effect of the coronavirus on the parathyroid gland (4). The aim of this review study is to discuss the possible involvement of the parathyroid gland in Corona virus disease-2019 (COVID -19) infection.

Materials and Methods

The previous documented literatures about the parathyroid and the involvement of parathyroid glands in COVID -19 patients were used for this review study.

Results and Discussion

The COVID-19 mainly causes pulmonary injury and severe hypoxic respiratory failure. The previous documented studies reported extrapulmonary expressions, gastrointestinal, neurological, cardiac, renal, cutaneous and ocular expressions of COVID -19. It was noticed that SARS-CoV was chiefly detected in lung, wind tube and pulmonary bronchus. Strangely it was noted in various structures and tissues including stomach, small intestine, sweat gland, pituitary, pancreas, adrenal, distal

convoluted renal tubule, parathyroid gland, liver and cerebrum (4,5).

The hypocalcemia is a usual laboratory outcome in viral diseases and pneumonia (6). The similar finding was noted by an another study in that nearly 65% of patients with novel COVID-19 had hypocalcaemia (7). It was commented that the severe COVID-19 diseased patients (62.6%) with hypocalcemia had a poor consequence and additionally hypocalcemia forecast a worse prognosis of severe COVID-19 infection (8). They notices that the advanced age, raised altitudes of CRP and IL-6 and hypocalcaemia were the threat features for COVID-19 patients and those are the bad prognostic elements.

The reason for decreased serum calcium would be vitamin D deficit, hypoalbuminemia, decreased bowel absorption of calcium and diminished secretion of and response to parathyroid hormone (PTH) secondary to augmented levels of inflammatory cytokines (7).

The PTH motivates the bony cells to discharge calcium by osteoclastic activity of the bones and also rises serum calcium by the reabsorption of calcium in the convoluted tube and at the loop of Henle of the nephron. The vitamin-D encourage the reabsorption of dietary calcium and phosphorus. Thus inadequate vitamin-D excites the parathyroid gland to discharge more PTH (8). There might be the situation of deteriorating of hypoparathyroidism during the COVID-19 infection and it was advised to deliver the vitamin-D to the Covid-19 patients. The vitamin-D is acquired naturally from the sunlight. The positive role of vitamin-D

replacement therapy in COVID-19 patients was noted by reducing the risk and severity of the COVID-19 disease (9).

Anatomically normal and abnormal parathyroid glands were identified by their size, form, consistency and histological judgments. Rounded edge, rise in consistency were noted in parathyroid adenoma whereas sharp edge, soft in consistency were noted in ordinary parathyroid gland (10).

There were documented studies mentioned about the association between the size of the parathyroid and PTH values (11,12). A previous study commented that the length of normal parathyroid gland ranged from 3 to 9 mm in the Bangladesh population. The glands of black populations were longer than white groups of people (13). The larger parathyroid gland secretes more PTH and the larger gland might have the greater number of chief cells (12,14). The size of the parathyroid gland literally means the total size of overall total number of parathyroid glands in an individual.

Microscopically the parathyroid glands have two cells, the chief cells secrete the parathyroid hormone (PTH) and the other one is the oxyphil cells and the detail action of oxyphil cell is not understood till now (15). The histologically normal glands have accumulation of intracellular fat which lies with the cluster of chief cells and those fats seem to be compressing the chief cells but in abnormal gland, the stromal fat appears to be compressed by parenchymal cells (10).

The quantity of fatty matters rise with aging and consist of almost 50% of the gland bulk in high aged population. Similarly more oxyphil

cells could be seen in elder people (16). Oxyphil cells present either as a single or in groups among the chief cells and they found to be bigger than chief cells and contain abundant huge mitochondria (16). The Oxyphil cells are absent in some species. Another type of cells named as transitional cells were noted in human parathyroid gland. Both transitional and oxyphil cells only answer and secrete the PTH to extensive period of stimulation of parathyroid gland (16). The involvement of the chief and oxyphil cells of parathyroid in COVID-19 patients in different ethnic groups need to be studied in detail.

The site of location of parathyroid was assessed by using neck ultrasonography and parathyroid scintigraphy. It was pointed out that enlarged parathyroid gland might be the reason for bone problem and not the result of bone disease (17).

It was documented that COVID-19 might upset the role of parathyroid glands by two likely ways. First is by directly performing on parathyroid tissues by SARS-Cov-2 virus and secondly by lung failure (7).

Conclusions

The size of parathyroid gland, the alteration in quantity of different cells of parathyroid in different age group, ethnicity, the serum calcium and vitamin-D level determines the amount of secretion of PTH in COVID-19.

Conflict of Interest

None declared

Corresponding Author

Romini Niranjan

Mobile Number :0779231686

E mail : rominiranjan@yahoo.com

CC BY 4.0

This is an Open Access article distributed under the terms of the Creative Commons Attribution 4.0 license (unless stated otherwise) which permits unrestricted use, distribution and reproduction in any medium, provided the original work is properly cited. Copyright is retained by the author(s).

**References**

1. Nanka, O., Sedy, J., Vitkovic, I., Libansky, P., Adamek, S., Prague. Surgical Anatomy of Parathyroid Glands with Emphasis on Parathyroidectomy, Medical Report. 2006;107(2): 261–272
2. Wang C. The anatomic basis of parathyroid surgery. Ann surg. 1976;183(3):271-275
3. Al-Bustami, F., and Khraisha, S. Variations in the Anatomical Position of the Normal and Diseased Parathyroid Glands. J Med J. 2009;43 (3):180-188
4. Elkattawy S, Alyacoub R, S. Ayad , M. Pandya , Ari Eckman . A Novel Case of Hypoparathyroidism Secondary to SARS-CoV-2 Infection. Cureus 2020;12(8): 1-6 e10097. DOI 10.7759/cureus.10097
5. Y. Ding, Li He, Q. Zhang, Z. Huang, X. Che, J. Hou, H. Wang, H. Shen, L. Qiu, Z. Li, J.Geng, J. Cai, H. Han, X Li, W. Kang, D. Weng, P. Liang and S. Jiang. Organ distribution of severe acute respiratory syndrome (SARS) associated coronavirus (SARS-CoV) in SARS patients: implications for pathogenesis and virus transmission pathways. J Pathol 2004; 203: 622–630
6. J. Liu, P. H. Jingwen Wu, J. Gong, D. Tian. Prevalence and predictive value of hypocalcemia in severe COVID-19 patients. Journal of Infection and Public Health 2020; 13:1224–1228
7. A.A., Alzwia. The effect of COVID-19 on parathyroid glands. Journal of Infection and Public Health. 2021; 14(6):724–725
8. A V. Pascalea, R.Finellia , R. Giannottia , V. Viscoa , D. Fabbriatorea , Ida Matulaa, P. Mazzeoa , N. Ragosab , A. Massaric , R. Izzod , E. Coscionic , M. Illarioe., M. Ciccarella, B. Trimarcod and G. Iaccarino. Vitamin D, parathyroid hormone and cardiovascular risk: the good, the bad and the ugly. Journal of Cardiovascular Medicine 2018;19(2): 62-66
9. Cutolo M, Paolino S, Smith V. Evidences for a protective role of vitamin D in

- COVID-19. RMD Open 2020;6:e001454. doi:10.1136/rmdopen-2020-001454
10. Yao, Kathy, Frederick R, Singer, Sanford, Roth, Aaron Aasoon, Ye, Cynthia and Armando E. Giuliano. Weight of Normal Parathyroid Glands in Patients with Parathyroid Adenomas. J Clin Endocrinol Metab, July 2004, 89(7):3208–3213 3211
 11. A. Ramas, A.Jakubovic-Cičkisić, S. Umihanic, M. Sulejmanovic, F. Brkic. Correlation between the Parathyroid Glands Size and Parathormones Value in Patients with Hyperparathyroidism. ORIGINAL PAPER | Med Arch. 2019; 73(4): 249-252 249
 12. L.Fang, B. Tang, D. Hou, M. Meng, M. Xiong and J. Yang. Relationship between parathyroid mass and parathyroid hormone level in hemodialysis patients with secondary hyperparathyroidism. Fang et al. BMC Nephrology 2015;16(82): 1-7
 13. Mahbub, S., Ara, S., Nurunnabi, A.S.M., Alim, A., Ahmed, R., Md. Alam, M.J., Nahar, Y. Morphological study of human parathyroid glands – A Postmortem Study. Bangladesh journal of Anatomy. 2013; 11(1): 15-18
 14. Ghandur, M.L., Cassady, J., Hajianpour, Paz, L., Reiss, E. The Parathyroid Gland in Health and Disease. AJP Parathyroid gland. 1986; 125 (2):.292-299
 15. Ritter, C.S., Haughey, B.H., Miller, B., Brown, A. J. Differential gene expression by oxyphil and chief cells of human parathyroid glands. J Clin Endocrinol Metab. 2012; 97(8): 1499–1505.
 16. H. Chen, T.Senda, S. Emura², Kin-ya Kubo. An Update on the Structure of the Parathyroid Gland. 2013 The Open Anatomy Journal, 2013; 5: 1-9
 17. Modarai, B., Sawyer, A., Ellis, H., CBE FRCS. The glands of Owen. J R Soc Med. 2004; 97:494–495

ORIGINAL RESEARCH

An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka

Chandimal KM¹, Sirak K², Edirisinghe EAST³, Adikari G⁴, Reich D², Yasawardene SG³

¹*Department of Anatomy, Faculty of Medicine, Wayamba University of Sri Lanka*

²*Department of Genetics, Harvard Medical School, USA*

³*Department of Anatomy, Faculty of Medical Sciences, University of Sri Jayewardenepura, Sri Lanka*

⁴*Postgraduate Institute of Archaeology, University of Kelaniya, Sri Lanka*

Abstract

Ancient DNA (aDNA) is used to explore the genetic landscape of the past and investigate the movement, mixture, and adaptation events that shaped present-day patterns of human genetic diversity. The present study was designed to explore the genetic composition of the ancient people of Sri Lanka. Despite the low amino acid racemization often found in skeletal remains of Sri Lanka, we attempted to extract DNA from skeletal remains of 5 individuals found from several sites in intermediate climatic zone in Sri Lanka by using methodologies particularly designed for highly damaged and degraded DNA. We attempted to extract and sequence DNA from the petrous part of the temporal bones and tooth roots of remains excavated from Sigiriya Potana, Pellemalala (4,500 YBP) and Miniatheliya (1,000 – 5,000 YBP) in Sri Lanka. The bone processing, DNA extraction and amplification were performed in three separate rooms with dedicated equipment as accordance to the recommendation for analysis of aDNA. All measures were strictly taken to minimize contamination with modern samples. Using the DNA extracts, we prepared dual-indexed single-stranded libraries treated with

uracil-DNA glycosylase (UDG) to reduce the rate of ancient DNA damage. Prior to sequencing, in-solution target hybridization was used to enrich the sequences that overlap the mitochondrial genome and about 1.24 million genome-wide SNPs. Sufficient data passing quality control standards for any of the five individuals studied was not obtained. Coverage of the mitochondrial genome ranged between 0.04-0.45x, with damage rates at the terminal nucleotide that were not indicative of authentic ancient DNA. A range of 453-2,838 SNPs out of ~1.24 million targets were covered across the nuclear genome, translating to a maximum coverage of 0.004x. The damage rates at the terminal nucleotide were not indicative of authentic ancient DNA. The low numbers of SNPs hit on the X and Y chromosomes precluded the confident assessment of genetic sex in all individuals but it is possible that the ancient skeletal sample represented by Sigiriya Potana is a female individual. Although the recent published work on using the petrous part of the temporal bone has generated many times more data than other skeletal elements, even this technique did not work to generate aDNA on this set of prehistoric skeletal remains found in the

Chandimal KM, Sirak K, Edirisinghe EAST, Adikari G, Reich D, Yasawardene SG - An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka

intermediate climatic zone in Sri Lanka. It is possible that other sites might provide better taphonomic preservation conditions than the sites we analysed, and in addition a priority should be to attempt ancient DNA analysis from skeletal remains found in Sri Lanka's dry zone.

Keywords: ancient DNA, prehistoric skeletal remains, Sri Lanka

Introduction

Ancient DNA (aDNA) is DNA from organisms that lived decades to hundreds of thousands of years ago. It can be used to explore the genetic landscape of the past and investigate the movement, mixture, and adaptation events that shaped present-day patterns of human genetic diversity. Ancient DNA can be recovered from biological remains (including hard and soft tissues) and has also been recovered from sediment as well as from residues on objects used by people; however, it is most common to extract DNA from bones and teeth. Particular elements of the human skeleton – including the petrous part of the temporal bone^{15,30}, tooth cementum^{2,11,18}, and the auditory ossicles³⁴ have been shown to preserve DNA much better than other skeletal elements (particularly elements of the postcranial skeleton but also other elements of the cranium and other parts of the teeth). The most frequently used skeletal element today in aDNA research is the petrous bone, and in particular the cochlea, which has shown to be even more DNA-rich than other parts of the petrous bone³⁰ (a protocol for

obtaining powder from the cochlea for aDNA analysis is published in Pinhasi, et al. 2019.)²⁹

While the identification and analysis of parts of the skeleton that have been shown to be particularly DNA-rich has greatly benefitted the field of aDNA research, allowing the recovery of greater amounts of DNA from contexts where bio-molecular preservation is likely to be poor, the DNA recovered from these skeletal elements is still invariably damaged because the enzymatic repair processes that maintained the molecules during life cease to function²⁴. At the time of an organism's death, endogenous nucleases begin to break down the nucleotide chain into small pieces that can be millions of times shorter than their original length, while hydrolytic and oxidation reactions fragment the DNA backbone and chemically modify the nucleotide bases^{5,19,24,27,28}. Post-mortem DNA damage accumulates at a rate that is influenced by various environmental factors, which include temperature, exposure to moisture, and the pH of the environment in which the remains were deposited or interred^{21,25,35}. DNA preserves best in cold and dry environments with stable temperatures, and worst in hot and humid places, particularly those that experience temperature fluctuations³⁵.

It is well known that the hot and humid environmental conditions in tropical environments promote DNA degradation and oftentimes results in poor DNA recovery from ancient biological materials^{3,8,9,31}. Although there is presently no reliable way to evaluate the preservation of DNA without carrying out DNA analysis, amino acid racemization has previously been used as a tool to estimate the

Chandimal KM, Sirak K, Edirisinghe EAST, Adikari G, Reich D, Yasawardene SG - An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka

preservation of DNA in bones^{8,31}. Reed (2003) reported that the preservation of a DNA is likely to be very poor in Sri Lanka according to records on low amino acid racemization of prehistoric bones found in highland dry caves³². Despite the low amino acid racemization found in skeletal remains of Sri Lanka, we attempted to extract DNA from skeletal remains found in intermediate climatic zones in Sri Lanka by using methodologies particularly designed for highly damaged and degraded DNA, as would be expected for remains from Sri Lanka.

On the basis of the annual rainfall pattern, Sri Lanka is classified into three main climatic zones: the wet zone, dry zone and intermediate zone. The wet zone runs over the southwestern region of the country which includes the central hill country and receives comparatively high mean annual rainfall of over 2,500 mm. The dry zone runs predominantly over the northern and eastern part of the country and receives a mean annual rainfall of less than 1,750 mm. The intermediate zone lies in between wet and dry zones and receives a mean annual rainfall between 1,750 to 2,500 mm, with a shorter and less-prominent dry season¹².

Prehistoric skeletal remains have been excavated from sites in different climatic zones in Sri Lanka. The sites of *Fa-Hien lina* at Bulathsinghala (34,000 ± 5,400 C¹⁴ YBP), *Batadomba lina* near Kuruwita (28,500-11,500 C¹⁴Y BP), *Beli lina* at Kitulgala (27,000 - 35,000 C¹⁴ YBP), and Alu lina at Attangoda - Kegalle (10,500 C¹⁴ YBP) are located in the wet zonal cave sites which have yielded human skeletal remains belonging to

Sri Lankan prehistory. *Bellan bandi palassa* at Balangoda (6,500 C¹⁴ YBP), located in the low country wet zone of the island, has also yielded human skeletal remains belonging to Sri Lankan prehistory²⁰.

The cave site of Sigiriya Potana, in the Matale district of Sri Lanka, which is situated in the intermediate zone, has yielded human skeletal remains dating to 4,000 YBP. Pellamalala which is one of the largest shell middens situated at Hambantota district in the Southern province in the intermediate zone has yielded human skeletal remains dating to 4,500 YBP. 36 Mini-atheliya, which is a shell midden area at the Hungama, located at Hambantota district, Southern province in the intermediate zone 23 has yielded human skeletal remains dated to 1,000 – 5,000 YBP.

The primary research question of our research was to explore the utility of ancient DNA technology to understand the genetic composition of the ancient people of Sri Lanka and in particular to investigate whether the present-day inhabitants of the Sigiriya region are genetically similar to these ancient people. Therefore, we attempted to extract and sequence DNA from the petrous part of the temporal bones and tooth roots of remains excavated from Sigiriya Potana, *Pellemalala* (4,500 YBP) and *Mini-atheliya* (1,000 – 5,000 YBP) in Sri Lanka. (Fig1.1).

Chandimal KM, Sirak K, Edirisinghe EAST, Adikari G, Reich D, Yasawardene SG - An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka

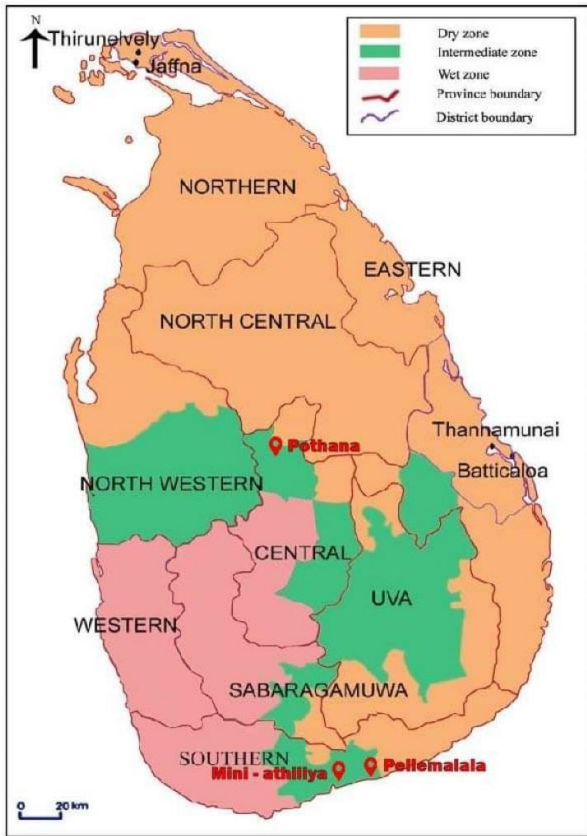


Fig 1.1 Location of the prehistoric sites from which remains were studied – *Potana*, *Pellemalala* and *Mini-ethiliya*. These sites are located in the intermediate zone in Sri Lanka.

Study Design, Materials and Methods

The present study was designed to extract and sequence DNA from human skeletal remains of five ancient individuals from sites within the intermediate zone in Sri Lanka and was carried out as a collaboration of researchers in Sri Lanka and the United States (Harvard Medical School - HMS; Boston, Massachusetts, USA). Included in the study were three individuals from the site of *Pallemalala* and one individual from the site of *Sigiriya Potana* who were curated at the Postgraduate Institute of Archaeology at University of Kelaniya, and one individual from *Mini-athiliya*, *Hungama*,

Hambanthota who was curated at the Star Fort Archaeological Museum at *Matara*. Ethical approval for this work was granted by the Ethics Review Committee of Faculty of Medical Sciences, University Sri Jayewardenepura, Sri Lanka. Two teeth that included tooth roots from *Pellemalala* and three petrous parts of skull fragments were transported to HMS for DNA analysis after obtaining official approval from the Director General of Archaeology in Sri Lanka for export of the remains and for their scientific analysis. The processing of skeletal remains and the extraction, amplification, sequencing, and analysis of DNA data was done at HMS.

The selection of particularly DNA-rich bone elements for analysis followed Pinhasi et al. 2015 and Hansen et al. 2017^{18,30}. The processing of bone material was carried out in a dedicated ancient DNA ‘clean room’ in order to minimize the impact of modern contamination. The powder for DNA extraction was produced from petrous samples (n=3) following the protocol described in Pinhasi (2019)²⁹ by combining the use of a Dremel disk saw and a fine sandblaster (Renfert Classic Basic). A drilling technique was used to create powder from the tooth roots (n=2). After processing the bone, DNA was extracted from ~50mg bone powder by dissolving it in a DNA extraction solution following published protocols^{10,22,33}. All bone processing was carried out by trained technicians wearing full cover suits, double gloves, hair nets, and face masks. All non-disposal equipment and work surfaces were sterilized through chemical cleaning as well as the use of UV-irradiation.

Chandimal KM, Sirak K, Edirisinghe EAST, Adikari G, Reich D, Yasawardene SG - An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka

From the DNA extracts, we prepared dual-indexed single-stranded libraries¹⁶ treated with uracil-DNA glycosylase (UDG) to reduce the rate of ancient DNA damage⁴. Prior to sequencing, we used in-solution target hybridization to enrich for sequences that overlap the mitochondrial genome and about 1.24 million genome-wide SNPs^{13,14,17,26}. We adopted this approach because we expected that these ancient samples would likely have low percentages of human DNA, making a strategy to enrich for informative positions more plausible than a shotgun sequencing approach. DNA libraries were sequenced using Illumina sequencing technology.

Results and Discussion

We did not obtain sufficient amounts of data passing quality control standards for any of the five individuals studied. Coverage of the mitochondrial genome ranged between 0.04-0.45x, with damage rates at the terminal nucleotide that were not indicative of authentic ancient DNA. A range of 453-2,838 SNPs out of ~1.24 million targets were covered across the nuclear genome, translating to a maximum coverage of 0.004x. For this small amount of DNA, the damage rates at the terminal nucleotide were not high enough to be indicative of authentic ancient DNA.

Although low numbers of SNPs hit on the X and Y chromosomes precluded the confident assessment of genetic sex in all individuals, the sequences from the extracted DNA from the ancient skeletal sample represented by Sigiriya *Potana* are consistent with being a female. Individual sex identification from skeletal

material has become a complimentary analysis to morphological assessment, the latter requiring at least some level of skeletal preservation and limited to adult individuals or sub-adults with soft tissue preservation. Sex determination is critical for investigating the demography of a population, such as sex-based population movements, marriage practices, settlement practices, and infanticide, among other topics. The female genetic sex of the *Potana* individual is consistent with morphological assessment by Chandimal et al. (2018)⁷. However, since there is no clear damage associated with the sequences, it is possible that the female DNA we have sequenced is not that of the ancient individual, but instead of a contaminating individual. An important aspect of ancient DNA research is to record the sex of the ancient individuals studied whenever possible in order to obtain greater insight into social structures or sex-biased practices in the past.

A likely reason that we failed to recover authentic aDNA from the five ancient individuals from Sri Lanka is that the bio-molecular preservation of these individuals was extremely poor due to the climatic features of Sri Lanka's intermediate zone. In addition, after removal from the archaeological site, samples are frequently stored in archives or museums, and the process of DNA degradation continues. Long term storage of archaeological specimens outside a lab freezer and prolonged exposure to UV irradiation can function to reduce the amount and the quality of DNA in ancient specimens⁶. This may be another factor contributing to the poor DNA preservation from of the individuals studied as part of this project. The skeletal samples of *Potana* and

Chandimal KM, Sirak K, Edirisinghe EAST, Adikari G, Reich D, Yasawardene SG - An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka

Pellamalala were excavated about ten years prior to their attempted DNA analysis, and are presently stored in boxes at room temperature at the museum and the osteology laboratory of the Postgraduate Institute of Archaeology, located in Sri Lanka wet zone where the environmental conditions are not conducive to the preservation of DNA.

As aridity, low temperature, and neutral or slightly alkaline pH are conditions that promote the preservation of DNA^{6,19}, a next step of this research project could be to attempt to recover DNA from skeletal remains found in Sri Lanka dry zone, such as Mantai, Jaffna, and Anuradhapura (among other sites), and also to attempt analysis of DNA from fresh excavations. Before attempting DNA analysis on such unique prehistoric bones, it would be informative to attempt DNA analysis on historic bones from the same or similar context, as it is evident that the DNA preservation in tropics is not ideal. Although the recent published work on using the petrous part of the temporal bone has generated many times more data than other skeletal elements, even this technique did not work out to generate aDNA on prehistoric skeletal remains found in intermediate climatic zone in Sri Lanka; however, given that the superior DNA preservation of this bone is evident, we believe that it should continue to be targeted in future ancient DNA work in Sri Lanka.

With the very limited availability of prehistoric skeletal remains it is of utmost importance that even negative results are published, and that researchers minimize the damage caused to each bone, recognizing that there is a non-trivial chance that their analyses will not yield

usable data based on current methods. Whenever possible, an antimeric should be preserved untouched for future research that may utilize more powerful methods than are available today. In addition, we encourage archaeologists to store human remains in temperature-controlled settings following excavation to reduce further degradation to aDNA within the skeletal remains.

Acknowledgement

The Department of Archaeology is acknowledged for granting approval for transportation of bone samples to the USA for aDNA analysis and Postgraduate Institute of Archaeology, University of Kelaniya is acknowledged for providing bone samples for the study. This work was supported by funds from the Howard Hughes Medical Institute and the John Templeton Foundation.

Conflict of Interest

None declared

Corresponding Author

Dr K M Chandimal
Department of Anatomy
University of Wayamba
Sri Lanka
kmchandimal@wyb.ac.lk

Chandimal KM, Sirak K, Edirisinghe EAST, Adikari G, Reich D, Yasawardene SG - An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka

CC BY 4.0

This is an Open Access article distributed under the terms of the Creative Commons Attribution 4.0 license (unless stated otherwise) which permits unrestricted use, distribution and reproduction in any medium, provided the original work is properly cited. Copyright is retained by the author(s).



References

1. Adikari and Adikari, G. 1998. Aspects of the prehistory of the Sigiriya-Dambulla Region. *Postgraduate Institute of Archaeology, Colombo*. (1998).
2. Adler, P.B., Seabloom, E.W., Borer, E.T., Hillebrand, H., Hautier, Y., Hector, A., Stanley Harpole, W., O'Halloran, L.R., Grace, J.B., Michael Anderson, T., Bakker, J.D., Biederman, L.A., Brown, C.S., Buckley, Y.M., Calabrese, L.B., Chu, C.J., Cleland, E.E., Collins, S.L., Cottingham, K.L., Crawley, M.J., Damschen, E.I., Davies, K.F., DeCrappeo, N.M., Fay, P.A., Firn, J., Frater, P., Gasarch, E.I., Gruner, D.S., Hagenah, N., Lambers, J.H.R., Humphries, H., Jin, V.L., Kay, A.D., Kirkman, K.P., Klein, J.A., Knops, J.M.H., La Pierre, K.J., Lambrinos, J.G., Li, W., MacDougall, A.S., McCulley, R.L., Melbourne, B.A., Mitchell, C.E., Moore, J.L., Morgan, J.W., Mortensen, B., Orrock, J.L., Prober, S.M., Pyke, D.A., Risch, A.C., Schuetz, M., Smith, M.D., Stevens, C.J., Sullivan, L.L., Wang, G., Wragg, P.D., Wright, J.P. and Yang, L.H. 2011. Productivity is a poor predictor of plant species richness. *Science*. 333, 6050 (2011), 1750–1753.
3. Bada, J.L., Wang, X.S. and Hamilton, H. 1999. Preservation of key biomolecules in the fossil record: Current knowledge and future challenges. *Philosophical Transactions of the Royal Society B: Biological Sciences*. 354, 1379 (1999), 77–87.
4. Briggs, A.W., Stenzel, U., Meyer, M., Krause, J., Kircher, M. and Pääbo, S. 2009. Removal of deaminated cytosines and detection of in vivo methylation in ancient DNA. *Nucleic Acids Research*. 38, 6 (2009), 1–12.
5. Brotherton, P., Endicott, P., Sanchez, J.J., Beaumont, M., Barnett, R., Austin, J. and Cooper, A. 2007. Novel high-resolution characterization of ancient DNA reveals C > U-type base modification events as the sole cause of post mortem miscoding lesions. *Nucleic Acids Research*. 35, 17 (2007), 5717–5728.
6. Burger, J., Hummel, S., Herrmann, B. and Henke, W. 1999. DNA preservation: A microsatellite-DNA study on ancient skeletal remains. *Electrophoresis*. 20, 8 (1999), 1722–1728.
7. Chandimal, K.M., Adhikari, G. and Yasawardene, S.G. 2018. Morphological Evaluation of Cephalic Phenotypes in

Chandimal KM, Sirak K, Edirisinghe EAST, Adikari G, Reich D, Yasawardene SG - An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka

- Purana (old) Inhabitants at Sigiriya Suburbs, Sri Lanka. *Sri Lanka Anatomy Journal*. 2, 2 (2018), 14.
8. Collins, M.J., Waite, E.R. and Van Duin, A.C.T. 1999. Predicting protein decomposition: The case of aspartic-acid racemization kinetics. *Philosophical Transactions of the Royal Society B: Biological Sciences*. 354, 1379 (1999), 51–64.
 9. Colson, I.B., Richards, M.B., Bailey, J.F., Sykes, B.C. and Hedges, R.E.M. 1997. DNA Analysis of Seven Human Skeletons Excavated from the Terp of Wijnaaldum. *Journal of Archaeological Science*. 24, 10 (1997), 911–917.
 10. Dabney, J., Knapp, M., Glocke, I., Gansauge, M.T., Weihmann, A., Nickel, B., Valdiosera, C., García, N., Pääbo, S., Arsuaga, J.L. and Meyer, M. 2013. Complete mitochondrial genome sequence of a Middle Pleistocene cave bear reconstructed from ultrashort DNA fragments. *Proceedings of the National Academy of Sciences of the United States of America*. 110, 39 (2013), 15758–15763.
 11. Damgaard, P.B., Margaryan, A., Schroeder, H., Orlando, L., Willerslev, E. and Allentoft, M.E. 2015. Improving access to endogenous DNA in ancient bones and teeth. *Scientific Reports*. 5, (2015), 1–12.
 12. Domros, M.Chandler, T.J. 1975. The Agroclimate of Ceylon. *Agricultural Meteorology*. 15, 1 (1975), 135–136.
 13. Fu, Q., Hajdinjak, M., Moldovan, O.T., Constantin, S., Mallick, S., Skoglund, P., Patterson, N., Rohland, N., Lazaridis, I., Nickel, B., Viola, B., Prüfer, K., Meyer, M., Kelso, J., Reich, D. and Pääbo, S. 2015. An early modern human from Romania with a recent Neanderthal ancestor. *Nature*. 524, 7564 (2015), 216–219.
 14. Fu, Q., Mittnik, A., Johnson, P.L.F., Bos, K., Lari, M., Bollongino, R., Sun, C., Giemsch, L., Schmitz, R., Burger, J., Ronchitelli, A.M., Martini, F., Cremonesi, R.G., Svoboda, J., Bauer, P., Caramelli, D., Castellano, S., Reich, D., Pääbo, S. and Krause, J. 2013. A revised timescale for human evolution based on ancient mitochondrial genomes. *Current Biology*. 23, 7 (2013), 553–559.
 15. Gamba, C., Jones, E.R., Teasdale, M.D., McLaughlin, R.L., Gonzalez-Fortes, G., Mattiangeli, V., Domboróczki, L., Kovári, I., Pap, I., Anders, A., Whittle, A., Dani, J., Raczky, P., Higham, T.F.G., Hofreiter, M., Bradley, D.G. and Pinhasi, R. 2014. Genome flux and stasis in a five millennium transect of European prehistory. *Nature Communications*. 5, (2014), 1–9.
 16. Gansauge, M.T., Aximu-Petri, A., Nagel, S. and Meyer, M. 2020. Manual and automated preparation of single-stranded DNA libraries for the sequencing of DNA from ancient biological remains and other sources of highly degraded DNA. *Nature Protocols*. 15, 8 (2020), 2279–2300.

Chandimal KM, Sirak K, Edirisinghe EAST, Adikari G, Reich D, Yasawardene SG - An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka

17. Haak, W., Lazaridis, I., Patterson, N., Rohland, N., Mallick, S., Llamas, B., Brandt, G., Nordenfelt, S., Harney, E., Stewardson, K., Fu, Q., Mittnik, A., Bánffy, E., Economou, C., Francken, M., Friederich, S., Pena, R.G., Hallgren, F., Khartanovich, V., Khokhlov, A., Kunst, M., Kuznetsov, P., Meller, H., Mochalov, O., Moiseyev, V., Nicklisch, N., Pichler, S.L., Risch, R., Rojo Guerra, M.A., Roth, C., Szécsényi-Nagy, A., Wahl, J., Meyer, M., Krause, J., Brown, D., Anthony, D., Cooper, A., Alt, K.W. and Reich, D. 2015. Massive migration from the steppe was a source for Indo-European languages in Europe. *Nature*. 522, 7555 (2015), 207–211.
18. Hansen, H.B., Damgaard, P.B., Margaryan, A., Stenderup, J., Lynnerup, N., Willerslev, E. and Allentoft, M.E. 2017. Comparing ancient DNA preservation in petrous bone and tooth cementum. *PLoS ONE*. 12, 1 (2017), 1–18.
19. Höss, M., Jaruga, P., Zastawny, T.H., Dizdaroğlu, M. and Pääbo, S. 1996. DNA damage and DNA sequence retrieval from ancient tissues. *Nucleic Acids Research*. 24, 7 (1996), 1304–1307.
20. Kennedy, K.A.R. 1993. Recent discoveries and studies of the human skeletal remains of ancient Sri Lanka, In P. K. Seth and S. Seth (eds). *New perspective in Anthropology*. New Delhi: MD publications. (1993), 299–320.
21. Kistler, L., Ware, R., Smith, O., Collins, M. and Allaby, R.G. 2017. A new model for ancient DNA decay based on paleogenomic meta-analysis. *Nucleic Acids Research*. 45, 11 (2017), 6310–6320.
22. Korlević, P., Gerber, T., Gansauge, M.T., Hajdinjak, M., Nagel, S., Aximu-Petri, A. and Meyer, M. 2015. Reducing microbial and human contamination in dna extractions from ancient bones and teeth. *BioTechniques*. 59, 2 (2015), 87–93.
23. Kulatilake, S. 2012. Early occupation of coastal Sri Lanka. In A.R. Sankhyan (eds). 16th Annual IUAES Conference Proceedings in Asian Perspectives on Human Evolution. (2012), 207–214.
24. Lindahl, T. 1993. Instability and decay of the primary structure of DNA. *Nature*. (1993), 709–715.
25. Lindahl, T. and Nyberg, B. 1972. Rate of Depurination of Native Deoxyribonucleic Acid. *Biochemistry*. 11, 19 (1972), 3610–3618.
26. Mathieson, I., Lazaridis, I., Rohland, N., Mallick, S., Patterson, N., Roodenberg, S.A., Harney, E., Stewardson, K., Fernandes, D., Novak, M., Sirak, K., Gamba, C., Jones, E.R., Llamas, B., Dryomov, S., Pickrell, J., Arsuaga, J.L., De Castro, J.M.B., Carbonell, E., Gerritsen, F., Khokhlov, A., Kuznetsov, P., Lozano, M., Meller, H., Mochalov, O., Moiseyev, V., Guerra, M.A.R., Roodenberg, J., Vergès, J.M., Krause, J., Cooper, A., Alt, K.W., Brown, D., Anthony, D., Lalueza-Fox, C., Haak, W., Pinhasi, R. and Reich, D. 2015. Genome-

Chandimal KM, Sirak K, Edirisinghe EAST, Adikari G, Reich D, Yasawardene SG - An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka

- wide patterns of selection in 230 ancient Eurasians. *Nature*. 528, 7583 (2015), 499–503.
27. O'Rourke, D.H., Hayes, M.G. and Carlyle, S.W. 2000. Ancient DNA studies in Physical Anthropology. *Annual Review of Anthropology*. (2000), 217–242.
 28. Paabo, S. 1989. Ancient DNA: Extraction, characterization, molecular cloning, and enzymatic amplification. *Proceedings of the National Academy of Sciences of the United States of America*. 86, 6 (1989), 1939–1943.
 29. Pinhasi, R., Fernandes, D., Sirak, K., Novak, M., Connell, S., Alpaslan-Roodenberg, S., Gerritsen, F., Moiseyev, V., Gromov, A., Raczky, P., Anders, A., Pietruszewsky, M., Rollefson, G., Jovanovic, M., Trinhhoang, H., Bar-Oz, G., Oxenham, M., Matsumura, H. and Hofreiter, M. 2015. Optimal ancient DNA yields from the inner ear part of the human petrous bone. *PLoS ONE*. 10, 6 (2015).
 30. Pinhasi, R., Fernandes, D.M., Sirak, K. and Cheronet, O. 2019. Isolating the human cochlea to generate bone powder for ancient DNA analysis. *Nature Protocols*. 14, 4 (2019), 1194–1205.
 31. Poinar, H.N. and Artur Stankiewicz, B. 1999. Protein preservation and DNA retrieval from ancient tissues. *Proceedings of the National Academy of Sciences of the United States of America*. 96, 15 (1999), 8426–8431.
 32. Reed, F.A., Kontanis, E.J., Kennedy, K.A.R. and Aquadro, C.F. 2003. Brief communication: Ancient DNA prospects from Sri Lankan highland dry caves support an emerging global pattern. *American Journal of Physical Anthropology*. 121, 2 (2003), 112–116.
 33. Rohland, N., Glocke, I., Aximu-Petri, A. and Meyer, M. 2018. Extraction of highly degraded DNA from ancient bones, teeth and sediments for high-throughput sequencing. *Nature Protocols*. 13, 11 (2018), 2447–2461.
 34. Sirak, K., Fernandes, D., Cheronet, O., Harney, E., Mah, M., Mallick, S., Rohland, N., Adamski, N., Broomandkshobacht, N., Callan, K., Candilio, F., Lawson, A.M., Mandl, K., Oppenheimer, J., Stewardson, K., Zalzal, F., Anders, A., Bartík, J., Coppa, A., Dashtseveg, T., Évinger, S., Farkaš, Z., Hajdu, T., Bayarsaikhan, J., McIntyre, L., Moiseyev, V., Okumura, M., Pap, I., Pietruszewsky, M., Raczky, P., Šefčáková, A., Soficaru, A., Szeniczey, T., Szoke, B.M., van Gerven, D., Vasilyev, S., Bell, L., Reich, D. and Pinhasi, R. 2020. Human auditory ossicles as an alternative optimal source of ancient DNA. *Genome Research*. 30, 3 (2020), 427–436.
 35. Smith, C.I., Chamberlain, A.T., Riley, M.S., Stringer, C. and Collins, M.J. 2003. The thermal history of human fossils and the likelihood of successful DNA amplification. *Journal of Human Evolution*. 45, 3 (2003), 203–217.

Chandimal KM, Sirak K, Edirisinghe EAST, Adikari G, Reich D, Yasawardene SG - An attempt to extract ancient DNA from the petrous part of the temporal bones and roots of teeth of skeletal remains found in the intermediate climatic zone in Sri Lanka

36. Somadeva, R. and Ranasinghe, S. 2006.
An Excavation of a Shell-midden at
Pallemalla in Southern Littoral area of Sri
Lanka: Some Evidence of Prehistoric
Chenier Occupation in c. 4th millennium
BC. *Ancient Asia*.

Morphometric Analysis of Dry Human Mandibles of Sri Lankans

Chandimal KM¹, Wickramrathna A¹, Wimalasinghe PD¹, Ranasinghe DM¹, Yasawardene SG²

¹Department of Anatomy, Faculty of Medicine Wayamba University of Sri Lanka, Kuliyaipitiya, Sri Lanka.

²Department of Anatomy, Faculty of Medical Sciences, University of Sri Jayewardenepura, Sri Lanka.

Abstract

Introduction: Variety of mandibular morphometric measurements determines the population and sex differences of facial skeleton. This study is aimed to determine the mandibular morphometric measurements of Sri Lankan population.

Materials and methods: Thirteen different mandibular measurements were obtained in 75 dry mandibles of which the gender and age were determined referring to the standard literature. Optimum measures were undertaken to reduce the intra and inter observer bias. The data were analysed using Minitab 19 version. Mean \pm SD, mean difference and P- value of each morphometric parameter for both sexes were analysed separately by using Minitab 19 version.

Results and discussion: The sample consisted of 40 male and 35 female mandibles, age ranging from 60 – 80 years with the mean age of 67.545 ± 4.36765 . The mean value of each morphometric parameter in males were higher except in the mandibular angle and the height and breadth of the mandibular body. The maximum mandibular length, bigonial width, maximum ramus height, bicondylar breadth, bicoronoid breadth, bimental breadth and mandibular notch depth of males showed statistical significance ($P < 0.05$). The mean

values of all the parameters were different from values of the different populations.

Conclusions: Mandibular morphometric characteristics of Sri Lankans showed distinctiveness within the population and a direct relationship with the sex. These findings provide information for objective assessment facial skeletons of Sri Lankans.

Keywords: Morphometry; human mandible; facial skeleton; Sri Lankans

Introduction

The lower jaw is formed by the mandible, which is the largest and strongest bone in the human face skeleton^{1,2,3,4,5}. The mandible, the largest bone in the face has horizontally curved body, convex forwards and two broad rami ascending posteriorly. The mandibular body is U shaped and it is formed by union of two left and right halves at the symphysis menti. The angle of the mandible is the place where the inferior margin of the mandible meets the posterior margin of the ramus. The mandibular rami which project perpendicularly upward contains head, neck and coronoid process^{2,3,4,5}

The gender, age, stature, life style, and health status of extinct individuals are all addressed in the detailed study of morphology and

Chandimal KM, Wickramrathna A, Wimalasinghe PD, Ranasinghe DM, Yasawardene SG - Morphometric Analysis of Dry Human Mandibles of Sri Lankans

morphometry of human skeletons^{2,6,7,8,9}. The mandible is one of the skull bones that can be used to establish a diseased person's gender, age, eating habits/diet, and oral health^{2,3,4,5,10,11}. The mandible is a compact bone that resists fragmentation and deterioration after death because it is a compact bone. The mandible is one of the most prevalent features in diseased individuals' skeletal remains, and it may be used to determine gender and estimate the age of the human at death because the jaw changes with time^{3,4,5,6,12}.

The sex determination of the victims found in warfare, tsunami, earth quake, landslides, explosions, forensic remains and archaeological remains is generally based on DNA studies and morphological and morphometrical assessment of pelvic bones and skull bones^{3,4,5,8,13,14}. When these bones are unavailable, sex determination of unknown are carried out by assessing mandibular morphometry and morphology such as gross size, robustness, gonion morphology, chin morphology, ramus morphology, muscular markings over the body surface of the mandible etc^{3,4,11,14,15}.

As mandible is the strongest bone of the face, there is less chances of its damage during disaster and accidents and even decaying after death. It was reported that the mandible remains its shape better than other bones for a long time after death of the individuals^{5,14,15,16}. This is of particular importance in relation to human identification as its durability. This quality was exploited to identify extinct individuals gender, age, facial morphologies etc and even determination of the ancestry of

the individual using available fragments of mandibles in archaeological skeletal remains by using mandibular dimensions as different populations across the world represented by different morphometric values^{5,6,17,14,16}.

Male and female mandibles differ from morphological features such as robustness, gonion morphology, chin morphology, ramus morphology, muscular markings over the body surface of the mandible^{3,4,11,14,15}. The morphological assessment of mandible of an individual is concluded with the individual's skills and experience and it depends on the person who takes part in the study and hence the morphological assessment becomes subjective and unreliable^{11,12,18,19}. Therefore, globally various morphometrical assessments on mandibles have been conducted to determine the variations of mandibular dimensions among gender in different populations in the globe^{20,21,22,23,24,25}.

These findings are more useful to maxillofacial surgeons, plastic surgeons, medico legal authorities, archaeologists and anthropologists for confirmation of their interpretation of mandibular observations in different populations and reconstruction of facial morphologies of the respective populations.

Researchers in different regions in the world are still working on dry or living mandibles to evaluate the morphology and morphometry of mandible and to analyse the relationship of the morphology and morphometry of the mandible to the particular population group and they concluded that different population show population specific morphometric parameters of the mandibles^{20,21,22,26,27,28}

Chandimal KM, Wickramrathna A, Wimalasinghe PD, Ranasinghe DM, Yasawardene SG - Morphometric Analysis of Dry Human Mandibles of Sri Lankans

Thus the present study was designed to determine the morphometrical variations of mandibles among male and female Sri Lankans.

Materials and Methods

The present study has been designed to analyse the morphometric parameters of male and female dry mandibles of Sri Lankans. The dry mandibles were collected from the Department of Anatomy, Faculty of Medicine, Wayamba University of Sri Lanka and Department of Anatomy, Faculty of Medicine, University of Peradeniya, Sri Lanka for the study. The fragmented mandibles and mandibles with abnormal morphologies were excluded from the study. The gender of the selected mandibles for the study was determined by assessing the morphological characteristics of the mandibles reported by Williams et al., 2000. The age of each mandible was determined by following the methods described by Williams et al., 2000. Thirteen mandibular measurements described in table 1.1 were taken from the 75 selected mandibles by following the bony landmarks of the diagrammatic representation (Fig 1.1). All the measurements were taken by using mandibular meter and digital Vernier calliper. Each measurement was taken thrice and mean value was taken for the analysis. Male and female mandibular measurements were analysed separately to generate morphometric parameters for male and female Sri Lankans.

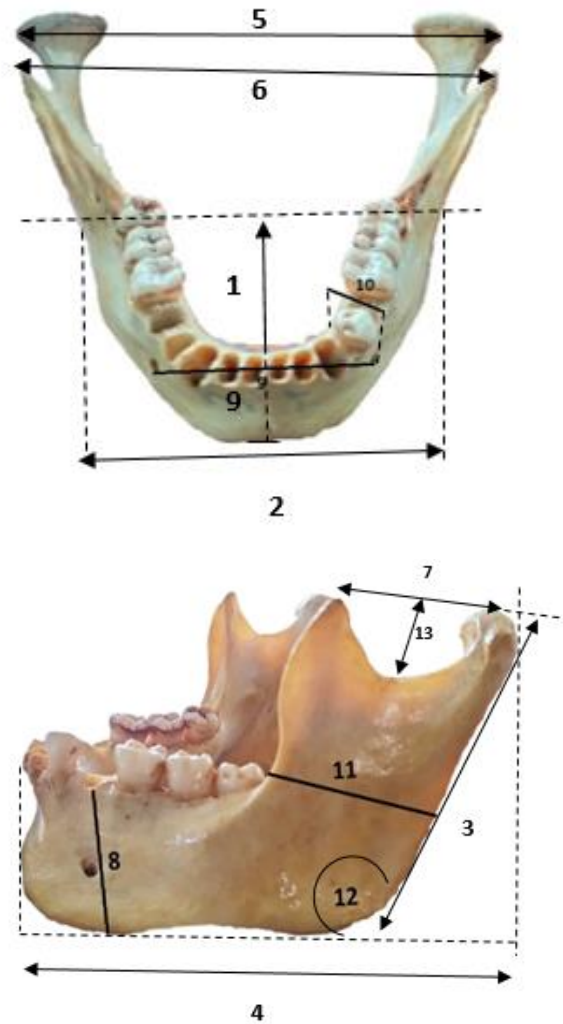


Figure 1.1. Mandible measurements

1-maximum mandibular length 2- bigonial width 3- maximum ramus height, right and left 4-mandibular length (projection) 5- bicondylar breadth 6- biconoid breadth 7- mandibular notch breadth 8- height of the mandibular body at the mental foramen 9- bimental breadth 10- breadth of the mandibular body 11- minimum ramus breadth 12- mandibular angle 13- mandibular notch depth

Chandimal KM, Wickramrathna A, Wimalasinghe PD, Ranasinghe DM, Yasawardene SG - Morphometric Analysis of Dry Human Mandibles of Sri Lankans

Table 1.1 Mandible measurements

| Mandibular measurements | Description of measurement using bony landmarks of mandible |
|-----------------------------------|--|
| 1- Maximum mandibular length | Distance from the anterior midline point on the chin (pogonion) to a center point of the bigonion line. |
| 2- Bigonial width | Direct distance between the right and left gonion. |
| 3- Maximum ramus height | Direct distance from the highest point on the mandibular condyle to the gonion. |
| 4- Mandibular length (projection) | Distance from the anterior midline point on the chin (pogonion) to the perpendicular line tangent to the posterior point of the left condyle. |
| 5- Bicondylar breadth | Direct distance between the most lateral points on the two condyles (condylion laterale). |
| 6- Biconoid breadth | Direct distance between the points at the tip of the two coronoid processes (coronion) |
| 7- Mandibular notch breadth | Direct distance from the condylion superior point to the coronion. |
| 8- Height of mandibular body | Direct distance from the alveolar process to the inferior border of the mandible perpendicular to the base at the level of the mental foramen. |
| 9- Bimental breadth | Direct distance between the most inferior point on the margin of the mandibular mental foramen (mentale). |
| 10- Breadth of mandibular body | Maximum breadth measured in the region of the mental foramen perpendicular to the long axis of the mandibular body |
| 11- Minimum ramus breadth | Minimum breadth of the mandibular ramus measured perpendicular to the height of the ramus |
| 12- Mandibular angle | Angle formed by the inferior border of the corpus and the posterior border of the ramus. |
| 13- Mandibular notch depth | Distance from the deepest part of the mandibular notch, to a center point of the condylion superior – (coronion) line. |

Mean \pm SD, mean difference and P- value of each morphometric parameter for both sexes were analysed separately by using Minitab 19 version. Sexual difference for each morphometric parameter was analysed with reference to p values. If the morphometric parameter's P value was less than 0.05, the parameter was substantially different between genders.

Chandimal KM, Wickramrathna A, Wimalasinghe PD, Ranasinghe DM, Yasawardene SG - Morphometric Analysis of Dry Human Mandibles of Sri Lankans

Results and Analysis

Forty male and 35 female mandibles were identified from 75 samples of mandibles. The age of the studied mandibles ranges from 60 – 80 years and mean age of the studied samples was 67.545 ± 4.36765 . In the present study thirteen measurements were taken from male and female mandibles as described in the material and methods. The mean, standard deviation, mean difference and P value for each measurement of male and female are given in the table 1.

Table 1.2 Mean \pm SD, mean difference and P- value of mandibular measurements among male and female Sri Lankans

| N = 75 | Sex | Mean | SD | Mean Difference | p-value |
|-----------------------------------|--------|----------|----------------|-----------------|---------|
| 1.Maximum mandibular length (mm) | Male | 82.3337 | ± 6.63680 | 3.89229 | 0.016 |
| | Female | 78.4414 | ± 5.36765 | | |
| 2.Bigonial width(mm) | Male | 96.2799 | ± 6.01647 | 6.30268 | 0.018 |
| | Female | 89.9772 | ± 12.41491 | | |
| 3.Maximum ramus height(mm) | Male | 57.8569 | ± 4.77926 | 3.40508 | 0.020 |
| | Female | 54.4518 | ± 6.00606 | | |
| 4.Mandibular length(mm) | Male | 112.4155 | ± 9.74738 | 11.34182 | 0.001 |
| | Female | 101.0737 | ± 13.38091 | | |
| 5.Bicondylar breadth(mm) | Male | 118.3070 | ± 3.62744 | 8.86046 | 0.000 |
| | Female | 109.4466 | ± 3.57986 | | |
| 6.Bicoronoid breadth(mm) | Male | 96.2194 | ± 5.52801 | 4.27446 | 0.002 |
| | Female | 91.9449 | ± 4.29034 | | |
| 7. Mandibular notch breadth(mm) | Male | 34.4180 | ± 2.91490 | 1.28335 | 0.118 |
| | Female | 33.1346 | ± 3.26412 | | |
| 8. Height of mandibular body(mm) | Male | 26.6168 | ± 5.46892 | -0.13913 | 0.909 |
| | Female | 26.7559 | ± 3.76178 | | |
| 9.Bimental breadth(mm) | Male | 45.1018 | ± 1.72666 | 1.01899 | 0.038 |
| | Female | 44.0828 | ± 1.94414 | | |
| 10.Breadth of mandibular body(mm) | Male | 10.8167 | ± 2.02543 | -0.04925 | 0.910 |
| | Female | 10.8659 | ± 1.26282 | | |
| 11.Minimum ramus breadth(mm) (R) | Male | 31.7475 | ± 2.74973 | 0.78997 | 0.250 |
| | Female | 30.9575 | ± 2.46941 | | |
| 12.Mandibular angle (°) | Male | 125.5718 | ± 7.58553 | -1.05219 | 0.621 |
| | Female | 126.6240 | ± 8.55523 | | |
| 13.Mandibular notch depth(mm) | Male | 14.5824 | ± 1.94141 | 2.17786 | 0.000 |
| | Female | 12.4045 | ± 1.67448 | | |

The mean mandibular measurements of males are higher than the female values except height and breadth of the mandibular body and mandibular angle. The maximum mandibular length, bigonial width, maximum ramus height, bicondylar breadth, bicoronoid breadth, bimental breadth and mandibular notch depth of males were significantly higher (<0.05) than the females.

Discussion

In this study, all mandibular measurements were obtained and documented using conventional literature, and male and female mandibles were compared. Except for height, breadth of the mandibular body, and mandibular angle, the results of this study revealed that males have higher morphometric parameters than females. Males had greater maximum mandibular length, bigonial width, maximum ramus height, bicondylar breadth, bicoronoid breadth, bimental breadth, and mandibular notch depth and breadth than females, and these morphometric parameters were substantially different between males and females.

According to the literature, it was reported that male mandibular morphometric parameters are often higher than female mandibular morphometric parameters^{20,23,27,29,28}. The present findings of morphometric dimorphisms of mandibles in this study are equivalent to other studies' findings of mandibular dimorphism^{22,23,27}. Gender disparities were discovered in these morphometric characteristics, which were similar to those identified in a study of South African indigenous, Indian, and Thai populations^{22,23,27}. Maximum mandibular length, bigonial width, maximum ramus height, bicondylar breadth, mandibular length, bicoronoid breadth, bimental breadth and mandibular notch depth of males in this study higher than the females and these eight mandibular morphometric characteristics are varied significantly between male and female Sri Lankans. In general, the findings revealed that male Sri Lankan mandibles were larger

than female mandibles, which is consistent with previous researches that have found male mandibles to be larger than female mandibles^{20,22,27}.

Varying ethnic groups around the world have different morphometry and morphology in their mandibles^{20,21,22,27,28,30}. The mean morphometric parameters of male and female Sri Lankans differ from those recorded for other populations around the world, such as Indians, Thais, and South Africans, as well as Brazilians^{20,22,27,31}. Males have a substantially greater bigonial width (96.28 mm) than females (89.98 mm) in this study, and the mean male and female values of bigonial width in this study are higher than those of Indians, who have a mean bigonial width of 79.76 mm for adult males and 73.83 mm for adult females²⁰. In this study, adult males had a bigonial width of 96.28 mm and adult females had a bigonial width of 89.97 mm, which is higher than the males' (83.20 mm) and females' (79.2 mm) recorded in the Thai population²⁷. The studied minimum ramus breadth for adult males and females in this study was 31.74mm and 30.95mm, respectively, which is larger than the Indians' minimum ramus breadth of 30.93mm for adult males and 29.57mm for adult females²⁰.

The reported Brazilian male and female mandibular morphometric values differ from the mandibular morphometric values of the Sri Lankans. Brazilian males and females have longer mandibular lengths (110.82 mm and 105.47 mm, respectively) than Sri Lankan males and females (82.33 mm) (78.44 mm). Bigonial widths are wider in Brazilian males (97.31 mm) and females (90.35 mm) than in

Chandimal KM, Wickramrathna A, Wimalasinghe PD, Ranasinghe DM, Yasawardene SG - Morphometric Analysis of Dry Human Mandibles of Sri Lankans

Sri Lankan males (96.28 mm) and females (90.35 mm) (89.98 mm). Brazilian males have a wider bicondylar width (119.10 mm) than Sri Lankan males (118.30 mm), and Brazilian ladies (114.68 mm) have a wider bicondylar width than Sri Lankan females (109.44 mm). The examined mandibular values of Sri Lankan males and females are, on average, lower than those reported for Brazilians³¹.

In adults, the angle of the mandible is around 140° ⁴, The mean angle of the mandible in adult males was $125.57^\circ \pm 7.58553$ in this study, and $126.62^\circ \pm 8.55523$ in females. The gender differences in the examined metrical angle of the mandible were compared to gender differences in the mean mandibular angle reported for young Indians ($119.92^\circ \pm 6.27^\circ$) and female ($125.20^\circ \pm 5.3^\circ$)²⁰. The mean mandibular angle was higher than that of Indian senior males ($124.13^\circ \pm 5.18^\circ$) and lower than that of females ($127.25^\circ \pm 7.46^\circ$)²⁰.

The mean angle of the mandible of females is larger than that of males, according to the literature^{32,33}, showed that the mean mandibular angle was greater in adult females (121°) than that in adult males (118°)³². The mean mandibular angle of Brazilian males (126.56°) and females (130.18°) were also in a same agreement that females are having larger angle of mandible than males. The gender differences of studied angle of mandible in this study (females > males) was compare with the Brazilians (females > males) and others too³¹. Although the mean value of the angle of mandible is higher in female than male in this study, the p-value was found to be greater than 0.05. Therefore, angle of mandible is insignificant among the two genders in Sri

Lankans. This is compared with reported study on Indians and Brazilians. The angle of mandible in Indians and Brazilians was insignificant among gender^{20,31}, Most of the recent studies showed that the angle of mandible has no correlation between the gender although the angle shows different in male and females^{32,33}.

The differences in muscular skeletal systems seen in males and females may account for the sexual dimorphism in mandibular morphometry and morphology. Because males have a more robust skeleton than females, muscles involved in the masticatory cycle adhere more strongly to the jaw, perhaps increasing the size and robustness of the mandible. Variable lifestyles and chewing habits may be linked to the size and form of the mandible. Male and female mandibles have different growth rates and developmental stages, according to research²⁶.

As reported in other investigations, there were discrepancies in the results of mandibular morphometry in this study. This could indicate that, aside from gender, different populations have distinct mandible sizes. To be able to apply the findings of this study to the best benefit, comprehensive studies must be undertaken and the mean difference morphometrical values for Sri Lankan male and female mandibles must be determined. The findings of this study will be useful in forensic medicine, forensic dentistry, anthropological investigations, and in the diagnosis and treatment planning of maxillo-facial surgeons and plastic surgeons, among other fields.

Chandimal KM, Wickramrathna A, Wimalasinghe PD, Ranasinghe DM, Yasawardene SG - Morphometric Analysis of Dry Human Mandibles of Sri Lankans

More research is needed, particularly to discover the morphological and morphometric variances in mandibles across the globe, as well as gender disparities. The morphometry of the mandible contributes significantly to the various shapes of the face, including facial angulation and chin prominence, which aids in the identification of persons belonging to various populations in various parts of the world.

Acknowledgements

The Department of Anatomy, Faculty of Medicine, Wayamba University of Sri Lanka and the Department of Anatomy, Faculty of Medicine, University of Peradeniya contributed dry mandibles for the study, which are gratefully acknowledged.

Conflict of Interest

None declared

Corresponding Author

Dr K M Chandimal
Department of Anatomy
University of Wayamba
Sri Lanka
kmchandimal@wyb.ac.lk

CC BY 4.0

This is an Open Access article distributed under the terms of the Creative Commons Attribution 4.0 license (unless stated otherwise) which permits unrestricted use, distribution and reproduction in any medium, provided the original work is properly cited. Copyright is retained by the author(s).



References

1. Dutta A. *Essential of Human Anatomy: Head and Neck*. In: *Essential of Human Anatomy*. 5th ed. Current Book International Calcutta; 1994. p. 40–4.
2. Lockhart R. *Anatomy of human skeleton*. In: Arnold E, editor. 2nd ed. London; 1965. p. 52.
3. Horan F. *Gray's Anatomy: the anatomical basis of clinical practice*. J Bone Joint Surg Br. 40th ed. 2008;91-B(7):983–983.
4. Williams PL, Bannister LG BM. *Gray's Anatomy*. 38th ed. Newyork: Churchill Livingstone; 2000. 409 – 419 p.
5. Bass WM. *Human Osteology A Laboratory and field manual*. 1979.
6. Humphrey LT, Dean MC, Stringer CB. *Morphological variation in great ape and modern human mandibles*. J Anat. 1999;195(4):491–513.

Chandimal KM, Wickramathna A, Wimalasinghe PD, Ranasinghe DM, Yasawardene SG - Morphometric Analysis of Dry Human Mandibles of Sri Lankans

7. Naccarato, Shawn; Petersen, Steven; John GL. *Skull features as clues to age, sex, race, and life style*. J Forensic Identif. 2008;58(2):172–81.
8. Jayachandra Pillai T, Sobha Devi T, Lakshmi Devi CK. *Bigonial dimeter, (11) Bicondylar diameter, (12) Bimental breadth, (13) Mandibular angle, (14) Length of lower jaw, (15) Interincisor width, (16) Interprmolal width*. 2014;13(1):8–15.
9. Inderbir Singh. *Textbook of Human Osteology*. 3rd editio. Jaypee Brothers Medical Publishers (P) Ltd.; 2006. 198–203 p.
10. Frayer DW, Wolpoff MH. *Sexual Dimorphism*. Annu Rev Anthropol. 1985;14(1):429–73.
11. Heeresh, C, Malaviya G. *The sexing of human mandible*. J Indian Acad Forensic Sci. 1972;2(2):7.
12. Hu KS, Koh KS, Han SH, Shin KJ, Kim HJ. *Sex determination using nonmetric characteristics of the mandible in Koreans*. J Forensic Sci. 2006;51(6):1376–82.
13. Rosa MA, Reimers EG, Fregel R, Vazquez JV, Darias TD, Gonzalez MA LJ. *Canary Island aborigine sex determination based on mandible parameters contrasted by amelogenin analysis*. J Archaeol Sci. 2006;
14. Scheuer L. *Application of osteology to forensic medicine*. Clin Anat. 2002;15(4):297–312.
15. Singh I. *Text book of human osteology*. 3rd ed. New Delhi: Jay Pee Brothers Medical Publisher; 2009. 198–203 p.
16. Vodanović M, Dumanović J. *Determination of Sex by Discriminant Function Analysis of Mandibles From two Croatian Archaeological Sites*.
17. Kaifu Y. *Changes in mandibular morphology from the Jomon to modern periods in eastern Japan*. Am J Phys Anthropol. 1997;104(2):227–43.
18. Loth SR, Henneberg M. *Mandibular ramus flexure: A new morphologic indicator of sexual dimorphism in the human skeleton*. Am J Phys Anthropol. 1996;99(3):473–85.
19. Tanveer Ahamed Khan H.S J. S. *Observation on morphological features of human mandibles in 200 South Indian subjects*. Anat Karnataka. 2011;1(5):44–9.
20. Sharma M, Gorea RK, Gorea A, Abuderman A. *A morphometric study of the human mandible in the Indian population for sex determination*. Egypt J Forensic Sci. 2016;6(2):165–9.
21. Fabian F, Mpembeni R. *Sexual dimorphism in the mandibles of a homogenous black population of Tanzania*. Vol. 28, Tanzania Journal of Science. 2004.
22. Franklin D, O’Higgins P, Oxnard CE. *Sexual dimorphism in the mandible of indigenous South Africans: A geometric*

Chandimal KM, Wickramrathna A, Wimalasinghe PD, Ranasinghe DM, Yasawardene SG - Morphometric Analysis of Dry Human Mandibles of Sri Lankans

- morphometric approach. S Afr J Sci.* 2008;104(3-4):101-6.
23. Reale M, Wilson GT. *A biometric study of one hundred and ten asiatic mandibles.* 2014;89(2):457-61.
24. Hoque MM, Ara S, Begum S, Kamal AM, Sayeed S. *Morphometric Analysis of Dry Adult Human Mandibular Ramus.* Bangladesh J Anat. 2014;12(1):14-6.
25. Morant AGM, Collett M, Adyanthaya NK, Morant BYGM. *Biometrika Trust A Biometric Study of the Human Mandible A biometric study of the human mandible and N. K. Adyanthaya,* 2014;28(1):84-122.
26. Franklin D, Oxnard CE, O'Higgins P, Dadour I. *Sexual dimorphism in the subadult mandible: Quantification using geometric morphometrics.* J Forensic Sci. 2007;52(1):6-10.
27. Ongkana N, Sudwan P. *Gender Difference in Thai Mandibles Using Metric Analysis.* Chiang Mai Med J. 2009;48(2):43-8.
28. Singh R, Mishra SR, S, Passey J, Kumar P, Singh S, et al. *Sexual Dimorphism in Adult Human Mandible of North Indian Origin.* Forensic Med Anat Res. 2015;03(03):82-8.
29. Rosas A, Bastir M. *Geometric morphometric analysis of allometric variation in the mandibular morphology of the hominids of Atapuerca, Sima de los Huesos site.* Anat Rec - Part A Discov Mol Cell Evol Biol. 2004;278(2):551-60.
30. Thakur KC, Choudhary AK, Jain SK, Kumar L. *Racial Architecture of Human Mandible-an Anthropological Study.* J Evol Med Dent Sci. 2013;2(23):4177-88.
31. Lopez TT, Michel-Crosato E, Benedicto E de N, Paiva LAS de, Silva DCB, Biazevic MGH. *Accuracy of mandibular measurements of sexual dimorphism using stabilizer equipment.* Braz Oral Res. 2017 Jan;31:e1.
32. Rai R, Ranade AV, Prabhu LV, Pai MM, Madhyastha S KM. *A pilot study of the mandibular angle and ramus in Indian population.* Int J Morphol. 2007;25(2):353-6.
33. Keen JA. *A study of the angle of the mandible.* J Dent Res. 2015;2(24):77-86.

CASE REPORT

Isolated giant mesenteric vein and anomalous inferior mesenteric vein insertion in the presence of cirrhosis – A cadaveric case report

Jayarathna MJS¹, Udayakumara S¹, Dhanushkar R¹, Weerasinghe S¹, Amaratunga HA¹

¹Department of Anatomy, Faculty of Medicine, University of Peradeniya.

Running title: Variations in abdominal veins

Abstract

The inferior mesenteric vein is one of the tributaries of the portal vein which receives blood from the large intestine, sigmoid colon and the rectum. It usually connects with the splenic vein before draining into the portal vein. It may however show variations such as joining the superior mesenteric vein directly or to the confluence of the superior mesenteric vein and splenic vein. Varices of the inferior mesenteric vein can be seen in the presence of portal hypertension due to various reasons and this can be isolated or in association with other portosystemic collaterals.

We report a case of a giant isolated inferior mesenteric vein with an anomalous insertion. The inferior mesenteric vein was seen to be connected to the superior mesenteric vein before draining into the portal vein. Further, on the distal end it was seen to be connected to the left renal vein, which in turn had dilated and enlarged. The left testicular vein was observed draining in to the inferior mesenteric vein but was not enlarged. The superior mesenteric vein, splenic vein and portal vein were not enlarged or varicosed even though the liver showed features of cirrhosis with no obvious signs of splenic enlargement.

This report contributes to enhance the knowledge on a rare anatomical connection between portal system and the systemic circulation and will be important to surgeons operating in this area.

Keywords: Inferior mesenteric vein, Varices, Renal vein, Testicular vein, Anomalous insertion

Introduction

Inferior mesenteric vein (IMV) is one of the tributaries of the portal vein. It is a large vein that drains blood from the large intestine. It usually ascends on the posterior abdominal wall and terminates in the splenic vein (SV) behind the body of the pancreas and the SV joins the superior mesenteric vein (SMV) to form the portal vein (PV). IMV begins as the superior rectal vein at about the mid-point of the anal canal, then passes along the inferior mesenteric artery to end in the SV. It receives blood from the superior rectal veins, the sigmoid veins and the left colic vein (Snell, 2012), (Kaur et al., 2017)

In this report, we present a case of an unusual giant IMV with an anomalous insertion in the presence of cirrhosis observed during routine dissections at the Department of Anatomy, University of Peradeniya.

Jayarathna MJS, Udayakumara S, Dhanushkar R, Weerasinghe S, Amaratunga HA - Isolated giant mesenteric vein and anomalous inferior mesenteric vein insertion in the presence of cirrhosis – A cadaveric case report

Case Report

During routine dissections at the Department of Anatomy Peradeniya, a giant, IMV with an anomalous insertion was observed in a cadaver of an adult male of Sri Lankan origin.

There were no surgical scars on the abdomen and no signs of intra – abdominal surgery in the peritoneal cavity. The liver showed cirrhotic features while the IMV was enlarged with varicosities along the whole length. There was no obvious splenic enlargement.

At the proximal end, the IMV was seen to be joining the SMV and forming a common stem which in turn joined the SV, thus forming the PV. Figure 1 shows the confluence of IMV, SMV with the common stem and the SV. At the distal end, the IMV was directly connected to the left renal vein on its inferior aspect and thus the systemic circulation forming a venous arch. The superior rectal veins, sigmoid veins and left colic veins were observed to be draining into this venous arch along its course. The broadest diameter of IMV was 31.12 mm.

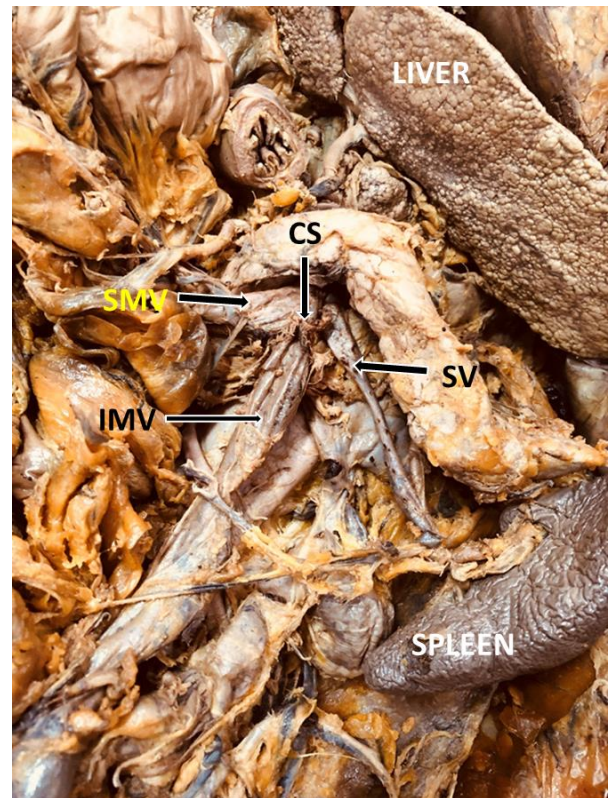


Figure 1: confluence of IMV, SMV to form common stem and confluence of common stem with SV to form portal vein.

(IMV – inferior mesenteric vein, SMV – superior mesenteric vein, SV – splenic vein, CS – common stem)

The left renal vein was also enlarged and dilated. The diameter of the left renal vein commencement at the inferior vena cava was 22.17 mm and at the confluence of IMV and renal vein it was 27.98 mm. Figure 2 shows the enlarged left renal vein, IMV and their anomalous connection.

Jayarathna MJS, Udayakumara S, Dhanushkar R, Weerasinghe S, Amaratunga HA - Isolated giant mesenteric vein and anomalous inferior mesenteric vein insertion in the presence of cirrhosis – A cadaveric case report

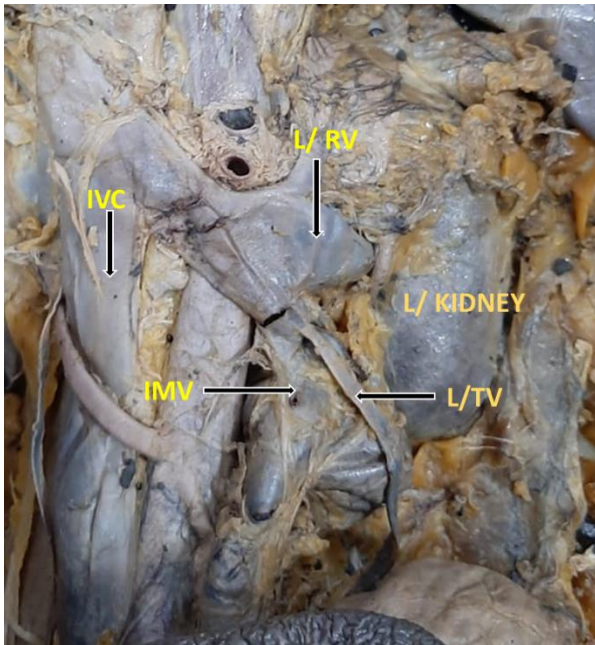


Figure 2: Enlarged left renal vein, IMV, left testicular vein and their anomalous insertion

(IVC – Inferior vena cava, L/RV – left renal vein, L/TV – left testicular vein, IMV – inferior mesenteric vein)

Figure 3 shows a diagram of the venous arch formed by the IMV.

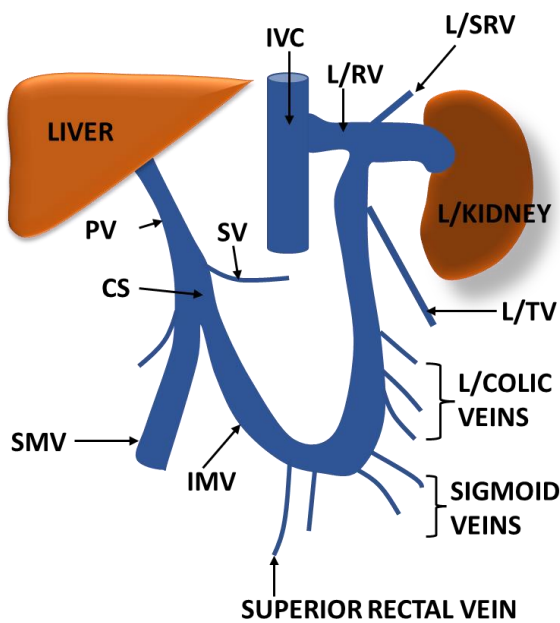


Figure 3: Diagram of venous arch formed by the inferior mesenteric vein and its connections

(IMV - inferior mesenteric vein, SMV – superior mesenteric vein, SV – splenic vein, PV – portal vein, CS – common stem, IVC – inferior vena cava, L/ RV – left renal vein, L/SRV – left suprarenal vein, L/ TV - left testicular vein)

The left testicular vein was observed draining in to the lateral side of the IMV just prior to its confluence with the left renal vein. The left testicular vein was not enlarged and its diameter was 4.69 mm. The SMV (diameter 7.56 mm), SV (diameter 7.56 mm) and PV (diameter 10.71 mm) were not enlarged.

Discussion

Cirrhosis of the liver is defined as the necrosis of liver cells followed by fibrosis and nodule formation (Feather, 2021). The end result is the impairment of liver function and gross distortion of the liver architecture leading to portal hypertension. An elevated pressure difference between systemic and portal circulation contributes to development of varices.

Liver cirrhosis accompanied by portal hypertension tend to form multiple portosystemic collaterals, between azygos and left gastric veins, between superior rectal vein and middle & inferior rectal veins, between portal tributaries of mesentery, mesocolon & retroperitoneal veins communicating with renal, lumbar & phrenic veins, between portal branches of the liver and the veins of the abdominal wall via veins passing along the falciform ligament from umbilicus, and between the portal branches in the liver and the

Jayarathna MJS, Udayakumara S, Dhanushkar R, Weerasinghe S, Amaratunga HA - Isolated giant mesenteric vein and anomalous inferior mesenteric vein insertion in the presence of cirrhosis – A cadaveric case report

veins of the diaphragm across the bare area of the liver (Feather, 2021), (Ellis, 2006).

Apart from these collaterals, IMV varicosities are also reported in the literature in the presence of portal hypertension due to numerous causes. Formation of varices on SMV is also reported. Further, large SMV varices communicating with the inferior vena cava through gonadal veins have also been reported (Akgul et al., 2003), (Federle & Clark, 1979), (Prasad et al., 2013).

Tributaries of IMV drains the large intestine, sigmoid colon and rectum. It has anatomical variations which include different drainage patterns. The more common variations include drainage in to the SMV and in to the confluence of SMV and SV (Kaur et al., 2017) (Graf et al., 1997)

The index case is unique with the IMV directly joining the left renal vein without collaterals, connecting the SMV and left renal vein and thus forming a venous arch. This venous arch has connected the portal and systemic circulations. Further, there is another portosystemic connection through the left testicular vein. As the SMV is not varicosed, this is a case of isolated IMV varix.

Acknowledgements

All the staff of department of Anatomy, Faculty of Medicine, University of Peradeniya.

Conflicts of Interests

The authors have no conflicts of interests.

Corresponding Author

Jayarathna M J S

32/2,

Kansagedarawaththa,

Kiribathkumbura

0778813089

Jayamini.jayarathna@med.pdn.ac.lk

Sri13485@gmail.com

CC BY 4.0

This is an Open Access article distributed under the terms of the Creative Commons Attribution 4.0 license (unless stated otherwise) which permits unrestricted use, distribution and reproduction in any medium, provided the original work is properly cited. Copyright is retained by the author(s).



References

1. Akgul, E. et al. (2003) "Giant varicose inferior mesenteric vein," *European Journal of Radiology Extra*, 45(3), pp. 122–125. Available at: [https://doi.org/10.1016/s1571-4675\(03\)00029-4](https://doi.org/10.1016/s1571-4675(03)00029-4). (Akgul et al., 2003)

Jayarathna MJS, Udayakumara S, Dhanushkar R, Weerasinghe S, Amaratunga HA - Isolated giant mesenteric vein and anomalous inferior mesenteric vein insertion in the presence of cirrhosis – A cadaveric case report

2. Ellis, H. (2006) in *Clinical anatomy: A revision and applied anatomy for clinical students*. 11th ed. Oxford: Blackwell Science, pp. 87–88. (Ellis, 2006)
3. Feather, A. (2021) *Kumar and Clark's Clinical Medicine E-Book*. 10th ed. Elsevier. (Feather, 2021)
4. Federle, M. and Clark, R.A. (1979) “Mesenteric varices: A source of mesosystemic shunts and gastrointestinal hemorrhage,” *Gastrointestinal Radiology*, 4(1), pp. 331–337. Available at: <https://doi.org/10.1007/bf01887551>. (Federle & Clark, 1979)
5. Graf, O. et al. (1997) “Anatomic variants of mesenteric veins: Depiction with helical CT venography,” *American Journal of Roentgenology*, 168(5), pp. 1209–1213. Available at: <https://doi.org/10.2214/ajr.168.5.9129413> (Graf et al., 1997)
6. Kaur, R. et al. (2017) “Variations in drainage pattern of inferior mesenteric vein,” *International Journal of Anatomy and Research*, 5(4.2), pp. 4581–4584. Available at: <https://doi.org/10.16965/ijar.2017.401>. (Kaur et al., 2017)
7. Prasad, G.R. et al. (2013) “Isolated inferior mesenteric portal hypertension with giant inferior mesenteric vein and anomalous inferior mesenteric vein insertion,” *Journal of Indian Association of Pediatric Surgeons*, 18(2), pp. 84–85. Available at: <https://doi.org/10.4103/0971-9261.109362>. (Prasad et al., 2013)
8. Snell, R., 2012. *Snell clinical anatomy by regions*. 9th ed. Philadelphia: Wolters Kluwer/Lippincott Williams & Wilkins, pp. 194-196. (Snell, 2012)

INSTRUCTIONS TO AUTHORS

The Sri Lanka Journal Anatomy publishes the following categories of articles which have relevance to Anatomy and allied sciences.

1. Leading articles - One article per issue. It may be solicited by the Editor. Authors are welcome to submit leading articles on current topics of interest. One's expertise or commentaries on general experiences etc. They should be approximately 1500 words in length. References should be 20 or less.
2. Reviews - Reviews are detailed surveys of published research pertinent to anatomy and associated sciences. They should be critical in nature and should not normally exceed 3000 words and 30 references.
3. Research articles - Articles resulting from research work belong to this group. Results from routine clinical examinations or laboratory investigations will not be considered under this category. They should not exceed 3000 words and 30 references. A reasonable number of tables and illustrations will be accepted.
4. Short reports - These include reports on current topics, modified techniques, new materials, practice management etc. Interesting results from routine work or laboratory investigations also may be accepted.
5. Case reports - Reports such as of rare anatomy related conditions.
6. Letters to Editors - Subjects unlimited, but may include short critique of published papers in the SLJA.
7. Miscellaneous topics - Subjects unlimited and the format are free. These may also include details of scientific meetings, conferences, annual sessions, examinations, news and views, visits and obituaries.
8. Proceedings of annual sessions - Abstracts from annual sessions of the ASSL and other colleges will be published under this category.

Submission of Manuscripts

Authors submitting a paper do so on the understanding that no part has been published before, that it is not being considered for publication elsewhere and that it has been read and approved by all the authors. Manuscripts including Tables and Figures should be sent in triplicate (hard copies) as the work will be reviewed by two or more referees. While papers are subject to editing the journal does not hold it responsible for statements made by the contributor. The author alone is responsible for the statements made in his paper. Submission of a manuscript means that authors automatically agree to assign exclusive copyright to the Anatomical Society of Sri Lanka if and when the manuscript is accepted for publication.

Manuscript on Disk

Once an article has been accepted for publication, the author will be asked to supply a copy of the final manuscript on disk (DVD) together with three hard copies of the complete manuscript. Every disk must be clearly labeled with the name of the author, title, software and program version number.

Manuscript Style

The manuscripts should be typed, double-spaced: on A4 (212x297 mm) paper and submitted in correct English: both British English and American spelling are acceptable, provided this is consistent throughout the manuscript. Manuscripts not submitted in proper format or in poor English may be returned without review. The format of a manuscript should be as follows: Title page, Abstract, Introduction, Material and Methods, Results, Discussion, Acknowledgements, References, Figure Legends and Tables arranged in that order.

Title Page –

The title page should contain the following information in the order given:

1. A concise but informative title
2. Author's full names (without degrees and titles)
3. Author's institutional affiliations

4. A running title not exceeding 40 letters and spaces
5. Name, address, telephone, telefax and electronic mail address of the author responsible for correspondence.

Abstract page –

Original and review articles must contain an abstract of approximately 250 words with four specified subtitles:

1. Objective: An introductory sentence indicating the objective and purpose of the study.
2. Material and methods: A description of experimental procedure including applicable statistical evaluation.
3. Results: A summary of the new; previous unpublished data and results.
4. Conclusion: A statement of the study's conclusion 3-5 key words according to Index Medicus should be provided.

Introduction –

The introduction should carry sufficient background information on the subject of study.

Material and Methods –

Procedures should be described in such detail as to make it possible to repeat the work.

Subheadings may be used to improve clearness. Correct unit abbreviations should be used (e.g. “h”, “min”, “s” and “Fm” rather than “hr”, “minutes”, “sec” and “FI” respectively). The authors should consider the ethical aspects of their research and ensure that the work has been approved by an appropriate Ethical Committee. Where applicable, a copy of the ethical clearance certificate should be attached. In human experimentation informed consent from individuals should be obtained and this should preferably be stated.

Statistical Analysis –

Since many scientific investigations rely on statistical treatment, authors are strongly urged to consult a person with in-depth statistical knowledge. Manuscripts with a clear element of statistics are regularly refereed by the Journal’s statistics consultant.

Results –

The results section should clearly and concisely present the findings of the research, as a rule in the past tense without subjective comments and reference to previous literature. The results should be supported by statistical or illustrative validation. For the sake of clarity this section may have subheadings.

Tables –

The tables should be numbered in the order of appearance in Arabic numerals. Each table

should have a brief explanatory title. Each table should be typed on a separate sheet, with due regard to the proportion of the printed column/page.

Figures –

All graphs, drawings, and photographs are considered figures and should be numbered in the order of appearance in Arabic numerals. Each figure should have a brief and specific legend, and all legends should be typed together on a separate sheet of paper. Photographs should be glossy prints and the reverse should give the figure number, title of paper, principal author’s name and have a mark indicating the top. Colour illustrations may be submitted in instances where their use may contribute significantly to the scientific value of the article. Colour illustrations may be printed free of charge at the Editor’s discretion, whereas others may be printed at the author’s expense.

References –

References are listed double-spaced in a separate reference section immediately following the text. References are numbered consecutively in the order in which they appear in the text; do not alphabetize. Identify references in texts, tables and legends by Arabic numerals (within parenthesis).

Congress abstracts should not be used as references nor may “unpublished observations” and “personal communications” be placed in the reference list. References cited

as “in press” must have been accepted for publication and not merely in preparation or submitted for publication.

Examples of correct forms of references are given below. These are based on the format used in the Index Medicus. Abbreviate journal names according to the /list of Journals Indexed, printed annually in the January issue of Index Medicus. List all authors; do not use et al in the reference list.

Journals

Standard journal article

Bartlett IG, O’Keefe P. The bacteriology of the perimandibular space infections. JOral Surg 1979; 37: 407-409. 64

Corporate (collective) author

WHO COLLABORATING CENTRE FOR ORAL PRECANCEROUS LESIONS. Definition of Leukoplakia and related lesions: an aid to studies on oral pre cancer. Oral Surg Oral Med Oral Pathol 1978; 46: 518-539.

Unpublished article Barker DS. Lucas RB. Localized fibrous growth of the oral mucosa. J Dent? Res 1965: in press.

Books and other monographs Pindborg JJ. Atlas of diseases of oral mucosa. 5th edition. Copenhagen: Munksguard, 1992: 50-66.

Chapter in book Boyde A. Amelogenesis and the structure of enamel. In: Cohen B. Kramer KH(eds). Scientific Foundations of Dentistry. William Heinemann Medical Books Ltd. London. 1976: 335-352.

No author given International statistical classification of diseases and related health problems, 10th revision, vol 1. Geneva: World Health Organization, 1992; 550-564.