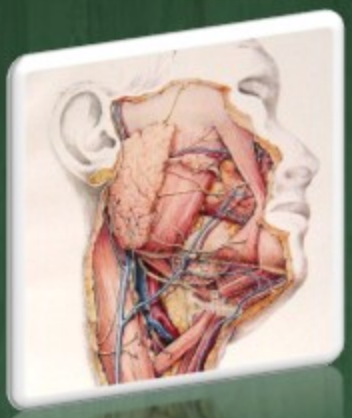
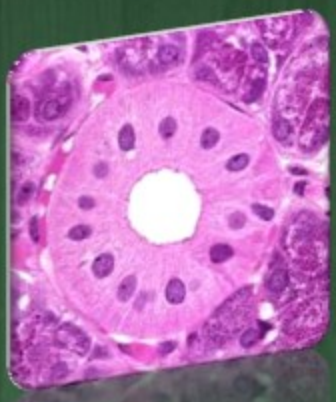
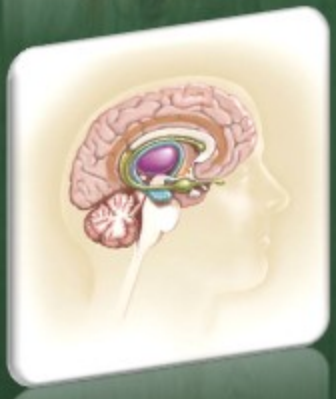
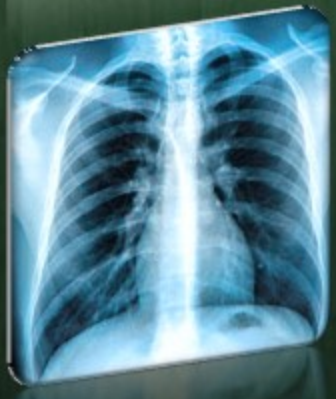
Volume: 31 (2), Dec 2023 ISSN: 0970 - 1842

JAS Journal of Anatomical Sciences

(U.P. Chapter of Anatomical Society of India)





Editor-in-Chief
Dr. Satyam Khare

Joint Editor
Dr. Shilpi Jain

www.asiupjas.com

About JAS

Journal Title	Journal of Anatomical Sciences
ISSN	0970-1842
E - ISSN	*(Application under process)*
Website	www.asiupjas.com
Email	asiupjas@gmail.com
Journal Categories	Gross & Comparative Anatomy, Histology & Histochemistry, Embryology, Neuroanatomy, Cytogenetics, Radiological Anatomy, Clinical Anatomy, Medical Education
Language	English
Inaugural Issue	December 1979
Frequency	Biannual (June - December)
Organisation	U. P. Chapter of the Anatomical Society of India
Editor-in-Chief	Dr. Satyam Khare (MS)
Joint Editor	Dr. Shilpi Jain (MD)
Associate Editors	Dr. Alok Tripathi Dr. Shobhit Raizaday
Current status	Active
Review process	Double-blinded peer review
Type of access	Open access
Full text format	PDF
License type	CC - BY (Creative Commons Attribution 3.0 licence)
Publication principles	International Committee of Medical Journal Editors (ICMJE)
Document Identifier type	DOI: 10.46351/jas
Editorial Office	Department of Anatomy Subharti Medical College Swami Vivekanand Subharti University Subharti Puram NH - 58, Delhi - Haridwar Bypass Road Meerut - 250005 Uttar Pradesh INDIA
Publisher	Dr. Satyam Khare *(for the U. P. Chapter of the Anatomical Society of India)*
Publisher Address	Department of Anatomy Subharti Medical College Swami Vivekanand Subharti University Subharti Puram NH - 58, Delhi - Haridwar Bypass Road Meerut - 250005 Uttar Pradesh INDIA Phone: 0121 – 3055000 (Extn: 2170)
Webmaster	Dr. Shobhit Raizaday *(Email: asiupjas@gmail.com)*

Instructions to Authors

CATEGORIES OF ARTICLES

The journal publishes –

- Original communications
- Brief communications including Case Reports
- Review articles
- Book reviews
- Scientific proceedings of U.P Chapter of the Anatomical Society of India

For original and brief communications, the journal accepts original research in the fields of –

- Gross anatomy and Comparative anatomy
- Embryology
- Histology and Histochemistry
- Cytogenetics
- Radiological anatomy
- Neuroanatomy
- Clinical anatomy
- Medical education

MANUSCRIPT PREPARATION

General Information –

- The manuscript should be written in British English and typed double-spaced throughout on **A4** paper size with 2.5 cm margin all around.
- All pages should be consecutively numbered in Arabic numerals.
- Sentences should not start with an abbreviation.
- The word *Figure* should be spelled out in the text except when in parenthesis: *example* – Figure 1 **or** (Figs. 2-3).
- All anatomical terms should be in conformation with those specified in Terminologia Anatomica (1998) Stuttgart.
- Nontechnical terms must be spelled according to the current Oxford English Dictionary.
- Numerical figures must be mentioned in Arabic numerals followed by abbreviated units in metric system.
- The manuscript should have a uniform style in a simple format. Complex formatting should be avoided.

Arrangement of the Main Document –
The main text of the manuscript should have the following subdivisions in sequence:

SEPARATE TITLE PAGE
TITLE & ABSTRACT
    INTRODUCTION
    MATERIALS AND METHODS
    RESULTS
    CONCLUSIONS (if any)
Text Body
    INTRODUCTION
    MATERIALS AND METHODS
    RESULTS
    DISCUSSION
    CONCLUSIONS
REFERENCES
Illustrations
Tables

(Start each subdivision on a new page)

SEPARATE TITLE PAGE

- The complete title of the paper.
- Full name of each author.
- List the affiliation of each author separately, linked to the author's name with a superscript number.

Example:
Rajveer Singh Chourasia[1], Ranjeet Kumar[2]
[1]Department of Anatomy, SLN Medical College, Koraput, Odisha, India
[2]Department of Anatomy, Hind Institute of Medical Sciences, Safedabad, Barabanki, UP, India

- Institution from which the paper emanated (with city, state and

zip/postal code)
- A short running title
- Address for correspondence

TITLE AND ABSTRACT
- Should be on the second page of each manuscript, not exceeding 300 words. The abstract should include a brief introduction, materials & methods, main results and important conclusions.
- A list of 3-9 key words should be given below the abstract.

TEXT BODY
- The manuscript text should be uniform in style and typed in Microsoft Word .doc or .docx format.
- The authors' name, affiliations or any other identifying information should NOT be included in the text body.
- The text of the manuscript should include, 1. Introduction, 2. Material and Methods, 3. Results, 4. Discussion, 5. Conclusion, if any, in that order.

REFERENCES
- References should be arranged chronologically as they appear in the body of the text according to the VANCOUVER SYSTEM.
- The reference number should be indicated in Arabic numerals in square brackets. *Example*: [1], [3-7], etc.

Papers published in periodicals:
1. Each author's surname followed by initials.
2. Full title of the paper
3. Abbreviation of the journal according to the style of index medicus
4. Year of Publication
5. Volume number, followed by a colon
6. First and the last page of paper.
Example: Longia, G.S., Kumar, V., and Gupta, C.D. Intra renal arterial pattern of human kidney -corrosion cast study. Ant. Anz., 1982., 166 :183-194.

Work referred from books:
1. Each Editor's surname followed by initials
2. Full title of the book
3. Name of the Chapter
4. Edition
5. Name of publisher
6. Domicile of publisher
7. Year of publication
8. Page numbers (a small 'p' with a full stop should be prefixed to the page number, pp with
a full stop if the number of the pages are more).
Example: Sinnatamby C.S. In Last's Anatomy Regional and Applied, Upper limb, llth ed., Churcill Livingstone London., 2005: p. 96.

Illustrations:
- All the illustrations including charts and diagrams and graphs should be referred to in the text and should be numbered.
- Every illustration should have legend typed on a separate page.
- For good reproduction, while printing, only good drawings and original photographs will be accepted.
- All the illustrations must be submitted in complete and finished form with adequate labelling.
- The abbreviations used in each illustration should be arranged alphabetically and should be included with the respective legends.

Tables:
- All the tables should be referred to in the text and should be numbered in Roman numerals.
- Every table should be prepared on a separate sheet and should have a heading (complete and brief) and may have footnotes.

- The tables should be simple with as few as possible vertical and horizontal lines.
- Too long and exhaustive tables are not acceptable because they can bot be reproduced as such.

ARTICLE PROCESSING CHARGES

Once accepted for publication, the authors are required to submit article processing charges for each submitted manuscript for the planning, scanning, plate making and printing on art paper as -

- @ Rs. 800/- (USD 25/-) per black & white photograph
- @ Rs. 1400/- (USD 40/-) per colored photograph
- @ Rs. 800/- (USD 25/-) per table, graph or drawn figures

If the total amount is less than Rs. 5000/-, it will be rounded off to a minimum of Rs. 5000/- (USD 150/-).

METHOD OF PAYMENT

Payment of the article processing charges are to be made via electronic funds transfer (RTGS/NEFT/IMPS) as per the following details.

Name of the Bank - ORIENTAL BANK OF COMMERCE
Branch - SKKB CHARITABLE TRUST M
Name of the Beneficiary - The Anatomical Society of UP Chapter
Account No. - 52282122034300
IFSC Code - ORBC0105228
MICR Code - 250022512

(Please submit the receipt of the transaction along with the manuscript with the first author's name as the file name of the receipt.)

For further instructions, please visit –

https://www.asiupjas.com/author-guidelines

https://doi.org/10.46351/jas.v31i2pp100-114

JAS Journal of Anatomical Sciences (U.P. Chapter of Anatomical Society of India)

Original Article

ASSOCIATION OF NUTRITIVE VALUE AND COGNITIVE PERFORMANCE IN ADOLESCENT GIRLS OF EASTERN UTTAR PRADESH

Durgesh Singh[1], Shashi Prabha Singh[1], Kshitij Raj[2], Mirza R U Beg[1], Devraj Yadav[3]

1. Department of Anatomy, Maa Vindhyavasini Autonomous State Medical College Mirzapur, India
2. Department of Community Medicine, Maa Vindhyavasini Autonomous State Medical College Mirzapur, India
3. Department of Paediatrics, Maa Vindhyavasini Autonomous State Medical College Mirzapur, India

ABSTRACT

Introduction: Food insecurity (FI) is a pressing concern affecting millions globally, particularly adolescents, impacting physical and cognitive health. Studies highlight its association with poor cognition, emphasizing the critical need for understanding and addressing this issue.

Materials and Methods: The study, conducted in urban slums of Saharanpur district, India, employed a community-based, cross-sectional design. Systematic random sampling was utilized, with 384 adolescent girls selected. Data collection included socio-demographic profiles, household food security assessment, cognitive performance evaluation, and morbidity analysis.

Results: Findings revealed a significant prevalence of morbid conditions among adolescent girls, including anemia, dental issues, and respiratory problems. Household food insecurity correlated with socio-demographic factors and significantly impacted cognitive performance, manifesting in high levels of anxiety, depression, and psychological distress.

Conclusions: Over 50% of adolescent girls in the study faced household food insecurity, with notable cognitive impairment and numerous morbidities. These results underscore the urgent need for public health interventions addressing food insecurity and socio-economic disparities, particularly in urban slum settings. Effective strategies should ensure access to nutritious food and promote socio-economic stability to mitigate the adverse effects of food insecurity on adolescent health and well-being.

Keywords : Cross-sectional study, Household food insecurity, Cognition, Adolescent girl, Urban slum, ICD 11

Address for Correspondence:
Dr. Durgesh Singh, Assistant Professor, Department of Anatomy, Maa Vindhyavasini Autonomous State Medical College Mirzapur, India
Email: dr.dpsingh84@gmail.com Mob: 9336404981

Date of Receiving: 19 Sep 2023
Date of Acceptance: 27 Oct 2023
ISSN: 0970-1842

INTRODUCTION

Food insecurity (FI) is the nutritionally capable ability to achieve the foods in culturally acceptable ways is narrow or ambiguous [1]. 79.5 crore people on earth face food insecurity [2], and FI is of swell care as less diet position changes the somatic and cognition of adolescents in the growing and mature world [3]. Studies on the correlation of food insecurity and poor cognition among adolescents describe that food insecurity will pace impaired cognition [4]. Studies of food deficient less gain of sufficient and nourishing nutriment article [5], decrease well-being, low functional health, shattered occupation, persistent health state, and increased depression and anguish [6]. Usual food instability is a mount people well-being complications of day-to-day less-earnings [7] and significantly positively identical with less employment and culture [8], large family size and food cost [9], and lower character of activity [10].

Cognition depicts a stage of happiness where the person knows to handle the normal stress of day-to-day happenings rapidly and is adept to make an addition to their own neighborhood [11]. The World Health Organization (WHO) described in the year 2014 almost 20% of adolescents go through poor cognition [12]. Adolescence is a critical cause of starting poor cognition, especially at an advanced juvenile stage [13]. Cognition in the midst of adolescents has a definite bond of presence at school, educational attainment, and eventual energy and freedom [14]. Also, poor cognition adds considerably to health concern deeds and capacity [15].

However, knowledge to describe the relationship in adolescent girls & habitat in slum areas [16]. The community that breathes in urban slums achieves impaired cognition; 34% living down to the starvation edge, 35% have no access to government fitness care machinery, and 17% interdepend on historic physicians for fitness care onset [17].

MATERIAL AND METHODS

Ethics: The study code of conduct was accepted by the Institutional Ethics Committee, MVASMC Mirzapur. The study desires and the agenda were distinctly told to every participant, and they were informed that they were free to withdraw at once without bias. Written informed consent was collected from all study participants. Privacy and confidence were applied all over the study period.

Study Design: The present study was a community-based, cross-sectional study conducted between September 2021 and October 2022 in 6 of 70 urban slum localities in Saharanpur district, India. This urban area is the habitat of slums that have grown continuously since the British era. About 38% of the community are urban poor and slum people, and 70 slums are assigned throughout the city [18].

Sampling: We used a laminated systemic random sampling design to select community-based samples. Initially, 6 slums were selected from 44 based on applications of slum grade laminated by people with probability proportion to size (PPS). As the second sampling stage, families were systematically selected from the slum profile, and girls were randomly chosen [2, 24, 40]. One adolescent girl from each family was selected using a random statistic table. For calculating the sample size, because of the non-availability of data about the prevalence of malnutrition in adolescent girls and varying prevalence of different disorders, the average prevalence of 50% for a confidence level of 95% with a relative precision of 10%, the sample size was 384. Anxiety, depression, loss of behavioral control, and psychological distress were calculated among food-secure and insecure groups.

Selection of Study Participants:

Inclusion criteria: Adolescent girls aged 13 to 19 years.

Exclusion criteria: Adolescent girls with mental illness such as intellectual disability, developmental delay, autism, or any other condition that inhibited communication or the ability to participate in the study were not included.

Data Collection: The survey consisted of: (1) personal profile (socio-demographic and socio-economic characteristics), (2) household food security, and (3) Cognitive performance. (4) Morbidity.

Food Insecurity: Food insecurity was measured with the Household Food Insecurity Access Scale (HFIAS), which divides participants into a) food-secure or b) mildly, c) moderately, or d) severely food-insecure over the previous 30 days based on the subject recall period. Defendants were interrogated to respond to questions with yes or no response based on their encounter during the previous 30 days. HFIAS scores were used as continuous measures of the degrees of family food insecurity and were calculated by adding outcomes for prevalence-of-event questions for each family. The highest outcome for a family was 27, i.e., the participant answered all nine prevalence-of-occurrence questions with a response code of 3 and the minimum score was 0, i.e., individuals answered “no” to all frequency-of-event questions. Thus, a higher outcome resulted in prominent food insecurity [19].

Cognitive Performance: Cognitive performance was measured with the PGI memory scale, which consisted of ten questionnaires and tasks to judge remote memory, recent memory, mental balance, attention & concentration, delayed recall, immediate recall, retention for a similar pair, retention for a dissimilar pair, visual retention, and recognition. The scale was based on a sketch-up of the last 30 days to go through

Cognitive performance	Level	Score	No of cases	Percentage
Anxiety	Low	9-24	6	1.5
	Medium	25-39	144	37.5
	High	40-54	268	69.79
Depression	Low	4-10	13	3.38
	Medium	11-16	164	42.7
	High	17-23	241	62.76
Loss of behavioral control	Low	9-22	1	0.26
	Medium	23-38	239	62.2
	High	39-53	178	46.3
Psychological distress	Low	24-60	1	0.26
	Medium	61-100	173	45
	High	105-142	244	63.5

Table 1. Cognitive performance of the study subjects

the ascent of anxiety, depression, behavioral control & psychological distress that further designed as low, medium, or high among adolescent girls aged ≥ 13 years. Moderate total memory scores (10 questionnaires, 15 marks each) were classified out of 150 as follows: (1) for anxiety, low 9–24, medium 25–39, and high 40–54; (2) for depression, low 4–10, medium 11–16, and high 17–23; (3) for loss of behavioral control, low 9–22, medium 23–38, and high 39–53; and (4) for psychological distress, low 24–60, medium 61–100, and high 105–142.

In our study findings for all components of cognitive performance, the proportion of participants logistic regressions were run (low vs. medium vs. high) as paired outcomes [20-21].

Morbidity: Physical examination revealed that adolescent girls were suffering from pediatric diseases, and morbidities were classified according to ICD-11 classification [22-23].

Socio demographic characters	Category	Food insecure	Food secure	Test of significance with p value
Age	13-16 years	107	92	x^2=4.65; df=1; *
	17-19 years	76	109	
Ethnicity	upper caste	13	30	x^2=0.23, df=2
	other backward caste	86	66	
	schedule caste	101	88	
Number of people in family	≤4	34	67	x^2 = 55.19; df = 4;***
	>4	123	160	
Number of siblings	≤2	30	65	x^2=19.18; df= 5; *
	>2	153	136	
Habitation in slum	≤30 years	178	193	x^2=27.22 ; df =3;***
	>30 years	5	8	
Education of subject	Primary and lower	98	154	x^2=6.89; df =2; *
	Secondary and upper	85	47	
Education of mother	Primary and lower	164	104	x^2=22.34; df =3; ***
	Secondary and upper	25	91	
Education of father	Primary and lower	104	66	x^2 = 92.46; df = 2; ***
	Secondary and upper	69	145	
Occupation of subjects	Student	140	193	x^2 = 28.87; df =1; ***
	Work outside	40	11	
Occupation of mother	Home maker	142	180	x^2 = 18.4; df = 1; ***
	Work outside	40	22	
Occupation of father	Agriculture/labor	88	170	x^2=41.36; df=2; ***,
	Service/business	70	56	
Family income	1st tercile	121	19	x^2=15.12; df=1; ***
	2nd tercile	40	78	
	3rd tercile	6	120	

Table 2. Associations between household food insecurity and socio-demographic characteristics

Statistical Analysis: Socio-demographic aspects were transformed in all multivariable logistic regression analyses to conclude the consequences of food insecurity on cognitive performance. Independent variables included in the multivariate analyses were: age, caste, family member, number of siblings, habitation in slum; education level; education of mother; education of father; occupation; occupation of mother; occupation of father; family income. Results were expressed as odds ratios (OR) & with 95% confidence intervals (CI) for paired conclusions of cognitive performance. All tests were two-tailed and p-values of <0.05 were treated as significant. Morbidities were conveyed with percentage. A statistical software IBM-SPSS version 17.0 was used for statistical analysis.

RESULTS

57.8% of adolescent girls were found to have one or more morbid conditions (Fig-1). 372 morbidities were found to be present in 222 sick girls, accounting for 1.67 morbidities per sick girl. Pallor was observed in 55.4%, Jaundice in 1.4%, Clubbing of fingers in 3.2%, Enlarged Lymph node in 8.1%, Hair problems in 15.3%, Angular stomatitis in 4.1%, Dental caries in 35.1%, Geographic tongue in 5.9%, Thyroid problems in 2.7%, Eye problems in 5.9%, Ear problems in 4.9%, Respiratory (throat + Tonsil) problems in 17.6%, and Skin problems in 8.1% (Table 5).

The prevalence per 1000 adolescent girls affected by disorders was determined according to ICD 11. The morbidity reported

Mental Health Status	Level	Food insecure	Food secure	Test of significance with p value
Anxiety	Low	1	5	x^2=75.91; df =2;***
	Medium	20	24	
	High	18	10	
Depression	Low	2	11	x^2 = 4.14; df=1; *
	Medium	25	21	
	High	32	26	
Loss of behavioral control	Low	1	0	x^2=38.85; df 1; ***
	Medium	33	36	
	High	20	18	
Psychological distress	Low	1	0	x^2=166.4; df 2; ***
	Medium	14	19	
	High	31	20	

Table 3. Association between household food insecurity and cognitive performance
p<0.05*,p<0.01*,p<0.001=significant**

	Characters	High anxiety	High depression	High loss of behavioural control	High psychological distress
		OR(95% CI)	OR(95% CI)	OR(95% CI)	OR(95% CI)
Food insecurity	No	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	Yes	17.129(7.414-39.572***	17.020(7.306-39.650) ***	8.35(5.29-13.16)	6.798(2.971-15.556) ***
Age	13-16 years	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	17-19 years	0.974(0.0.517-1.832)	0.048(0.027-0.086	0.277(0.135-3.572) **	0.048(0.027-0.086) ***
Ethnicity	upper caste	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	other backward caste	1.349(0.838-2.173)	1.787(1.040-3.069)*	1.591(0.932-2.718)	1.54(1.13-2.08)*
	schedule caste	1.49(1.00-2.23)*	2.55(1.40-4.65)**	1.56(1.19-2.06)***	1.75(1.13-2.72)*
Number of family member	≤4	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	>4	1.29(0.95-1.76)	1.84(1.16-2.93)	1.591(0.932-2.714)	1.370(0.849-2.209)
Number of siblings	≤2	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	>2	1.21(0.67-2.18)	1.96(1.08-3.53)*	1.15(0.49-2.7)	1.68(1.20-2.36)*
Habitation in slum	≤30 years	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	>30 years	3.64(2.17-6.08)***	2.22(1.45-3.40)**	1.97(1.36-2.87)**	1.52(1.15-2.01)*
Education of subject	Primary and lower	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	Secondary and above	1.582(0.932-2.684)	1.912(1.025-3.566)*	1.90(1.10-3.28)	1.69(1.20-2.36)
Education of mother	Primary and lower	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	Secondary and above	2.321(1.143-4.714)	3.08(1.67-8.52)	2.143(1.260-3.644)	2.29(1.53-3.43)
Education of father	Primary and lower	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	Secondary and above	2.063(1.121-3.799)	3.42(2.19-5.34)	3.22(1.78-5.82)**	2.15(1.26-3.68)**
Occupation of subjects	Student	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	Work outside	5.24(2.77-9.14)**	4.514(2.177-9.360)***	5.93(3.15-11.2)***	5.77(2.77-9.94)***
Occupation of mother	Home maker	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	Work outside	2.78(1.86-4.14)***	2.925(1.205-7.099)***	1.737(1.014-2.975)*	2.020(1.093-3.735)**
Occupation of father	Service/business	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	Agriculture/labour	2.778(1.149-6.718)***	3.746(1.743-8.049)**	3.511(1.9176.428)***	3.585(1.961-6.554)***
Family income	1st tercile	1.00(ref)	1.00(ref)	1.00(ref)	1.00(ref)
	2nd tercile	2.063(1.21-3.799)*	2.245(1.098-4.589)*	2.321(1.143-4.714)*	2.143(1.260-3.644)***
	3rd tercile	6.798(2.971-15.556)***	7.177(3.344-15.409)*	7.575(3.511-16.343)*	9.05(4.81-17.1)***

Table 4. Logistic regression analyses of relations between cognitive performance and household food insecurity and subject characteristics determined by multivariate analysis of odds ratio(OR)&CI 95%.

Tuberculosis at 7.5, Scabies at 35, Nutritional anaemia at 307.5, Disorders of the thyroid gland at 15, Disorders of the conjunctiva at 12.5, Disorders of the external and middle ear at 27.5, and Acute upper respiratory infections at 97.5.

DISCUSSION

The present study reported high anxiety at 69.79%; high depression at 62.76%; medium loss of behavior at 62.2%; and high psychological distress at 63.5%. A similar study reported high anxiety, depression, psychological distress, and loss of behavioral control [24]. Living in urban slum households with poor quality of life was suggested to influence the mental health status of adolescent girls [25].

The present study reported associations between household food insecurity and socio-demographic characteristics. A study reported that 50% suffered from food insecurity, and 4.5% of households suffered moderately and severely from food insecurity, which is higher than studies conducted in the USA, Canada, France, Ethiopia, and South Africa [30]. In the present study, family member ≤4 vs. >4, number of siblings ≤2 vs. >2, and habitation in the slum ≤30 vs. >30 reported significant similarities to a study conducted in an urban slum [23].

This study found significant associations between the mother's occupation (homemaker vs. work outside) and father's occupation (agriculture/labour vs. service/business) and family income (1st tercile vs. 2nd tercile vs. 3rd tercile) [24, 26]. All cognition factors showed significance compared with a low family income in the first and second terciles, being more likely to have mental health issues [24, 26].

In the present study, associations were found between cognitive performance and household food insecurity and subject characteristics for paired conclusions of cognitive performance. All cognition factors related to household food insecurity showed significance in slum habitation. Similarly, it was reported that those living in lower socioeconomic status, disabled individuals, and housewives living in slums are at risk of poor mental health status [27]. Adolescent girls' occupation (student vs. work outside) was significant. Job shock has an impact on mental health [27]. Furthermore, they must work as adults, and elevated mental health problems are frequent among employed adolescent girls [29].

In our study, cognitive factors such as anxiety, depression, loss of behavioral control, and psychological distress were significant. Except for high loss of behavioral control, all factors showed significance in

food insecurity. A study of multivariate analysis showed that household food insecurity was strongly associated with high anxiety, high depression, high loss of behavioral control, and high psychological distress, which are consistent with the results of many other studies [30]. Despite no occasional mechanism between HFI and negative cognition being settled, the confusion of food supplies maintenance enhances stresses that might contribute to low cognition.

More importantly, HFI may increase the risks of anemia, lower nutrient intakes, cognitive problems, aggression, and anxiety. Studies have shown that anemia affects cognition by its direct neurochemical effect and by its indirect effect on behavior, as individuals become less attentive and less responsive. The main pathogenesis may be iron, throughout the white matter, being more in the basal ganglia. Iron deficiency anemia affects cognition by causing a decrease in the iron concentration in the brain, hence reducing neurotransmitter levels leading to hypomyelination and delayed neuromaturation [41, 43].

High depression and high psychological distress factors showed significance in siblings, and only the high depressed factor showed significance in the education of subjects. High depression and high psychological distress in the Other Backward Class, and all factors in the Scheduled Caste, were significant, compared to similar studies, which also show adolescent girls from socio-economically disadvantaged groups (Scheduled Caste/Scheduled Tribe) [24].

In the present study, all factors were significant in the occupation of subjects and the occupations of the mother and father. A paper reported that socioeconomically disadvantaged adolescents with a persistent low socioeconomic status were more likely to develop mental health problems [33], and poverty has direct effects on adolescent mental health [26]. Subjects' education, mother's education, and father's education (primary & lower vs. secondary & upper) reported significant findings. Loss of behavioral control and psychological distress factors showed significance in the education of the father, compared to the study, where paternal education of primary or lower places teenage daughters at risk of mental health problems [24]. A study reported that fathers with a primary school level of education had significantly higher emotional and behavioral problem scores than fathers educated to higher levels [41].

In the present study, high loss of behavioral control and high psychological distress factors and the ratio of age 13-16 years vs. age 17-19 years reported significant findings compared to having lower levels of anxiety,

depression, and psychological distress than older teenage girls (17–19 years) [24]. Although most mental disorders begin between the ages of 12–24 years, they are often detected in later life [28]. A study reported that around half of lifelong mental disorders start before 14 years of age [29], and the Australian National Survey of Mental Health and Wellbeing (NSMHWB) found that 27% of 18- to 24-year-olds had mental disorders [42].

57.8% of adolescent girls were sick with one or more morbid conditions, and 372 types of morbidities were present in 222 sick girls, accounting for 1.67 morbidities per sick girl. Pallor was observed in 55.4%, dental caries in 35.1%, and respiratory problems in 17.6%. The prevalence of nutritional anemia was reported highest at 307.5 in adolescent girls compared to morbidity per 1000. Due to infection in the upper respiratory tract (97.5), diseases of the oral cavity (55), diseases of the skin including scabies (35), and the percentage of prevalence of diseases of the middle and external ear (27.5%), diseases of the thyroid (15%) were reported.

The study reported anemia (55.5%), dental caries (37.2%), pediculosis (31%), URTI (17.5%), refractory errors (13.4%), and acne (11%) [36]. Another study reported the prevalence of inadequate oral hygiene (55.4%), pediculosis (39.2%), cold & cough (25.8%), lymphadenopathy (22.2%), scabies (16.2%), inflamed tonsils (7.8%), fever (7.5%), and ear discharge (7%) among adolescent girls [32]. A study reported that 94.5% of girls had one or more morbid conditions, with pediculosis (87.5%), dental caries, and skin disorders (50% each), worm infestation (18.3%), ENT disorders (17.5%), clinical anemia (5.8%), and defective vision (4.7%) [37].

Based on these reports, importance should be given to the importance of early mediation to improve mental health outcomes in adolescents. Our results do not rule out the possibility that poor mental health status among adolescent girls causes household food insecurity [34].

Limitations: The study has several limitations. First, we did not compare the degrees of associations between adolescent girls from households with marginal, low, or very low food security and mental health, or between adolescent girls and household heads. Because this study was conducted on a homogenous population of adolescent girls living in slum areas, we were unable to compare relations between adolescent girls living in slum areas and non-slum areas or between male and female adolescents. Second, we conducted the study with a small sample size that might suffer from poor external validity. Third, in the statistical analyses, we treated low and medium food insecurity as one category. Fourth, because

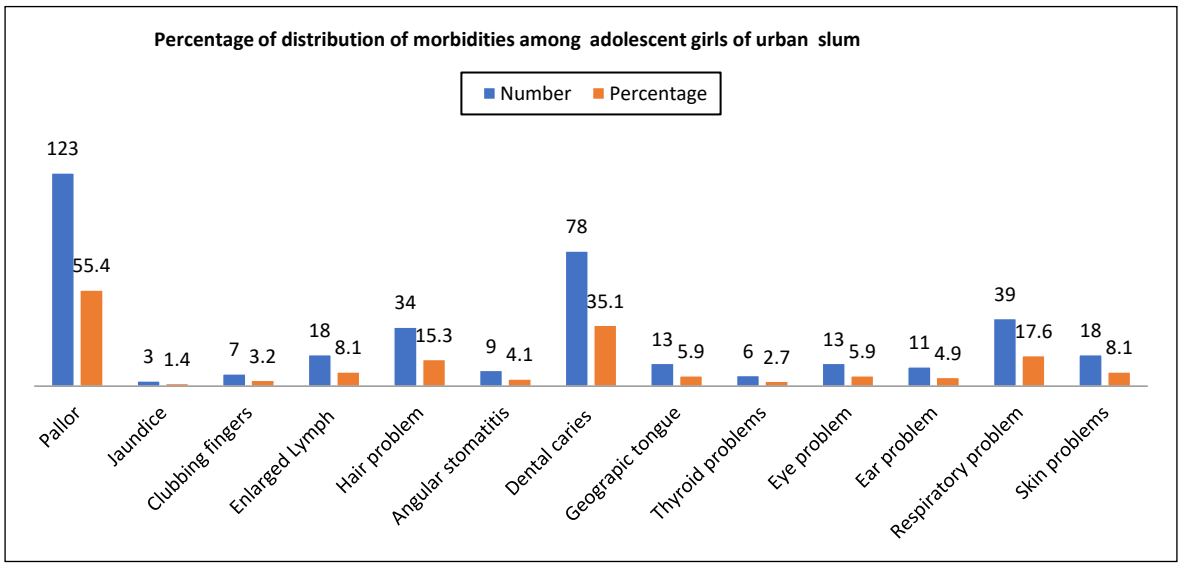


Fig. 1. Distribution of girls according to various morbidities

Code	Diseases	No.	Prevalence/1000
A15-A19	Tuberculosis	3	7.5
B85-B89	Scabies	14	35
D50-D53	Nutritional anaemias	123	307.5
E00-E07	Disorders of thyroid gland	6	15
H10-H13	Disorders of conjunctiva	5	12.5
H65-H75	Disorders of external and middle ear	11	27 .5
J00-J06	Acute upper respiratory infections	39	97.5
K00-K14	Diseases of oral cavity, geographic tongue, angular stomatitis	22	55

Table 6. Prevalence of morbidities among adolescent girls

of the cross-sectional nature of the study, we report only associations as we could not determine causalities. Finally, some potential confounders, such as exposure to violence, were not adjusted for in the present study, which may have impacted mental health outcomes [35].

CONCLUSION

Approximately more than 50% of adolescent girls were affected by household food insecurity and experienced cognitive impairment, anxiety, depression, psychological distress, and loss of behavioral control, with 372 types of morbidities present in 222 sick girls, accounting for 1.67 morbidities per sick girl. These results are much greater than those reported in the developed world. In our study, food insecurity was independently associated with impaired cognition, requiring clear-cut public health interventions that include access to tolerable, safe, and nutritious food and socio-economic changes to be implemented. Food insecurity in Indian slums should be addressed by specific public health intervention programs that provide access to satisfactory, nutritious food.

REFERENCES

1. Anderson SA. Core indicators of nutritional state for difficult-to-sample populations. J Nutr. 1990;120:1555-1600.
2. Food and Agriculture Organization; International Fund for Agricultural Development and World Food Program. The State of Food Insecurity in the World 2014. Strengthening the Enabling Environment for Food Security and Nutrition. Rome, Italy: Food and Agriculture Organization; 2014.
3. Eicher-Miller HA, Zhao Y. Evidence for the age-specific relationship of food insecurity and key dietary outcomes among U.S. children and adolescents. Nutr Res Rev. 2018;10:1-16.
4. Pryor L, Lioret S, van der Waerden J, Fombonne E, Falissard B, Melchior M. Food insecurity and mental health problems among a community sample of young adults. Soc Psychiatry Psychiatr Epidemiol. 2016;51:1073-1081.
5. Bocquier A, Vieux F, Lioret S, Dubuisson C, Caillavet F, Darmon N. Socio-economic characteristics, living conditions and diet quality are associated with food insecurity in France. Public Health Nutr. 2015;18:2952-2961.
6. Vozoris NT, Tarasuk VS. Household food insufficiency is associated with poorer health. J Nutr. 2003;133:120-126.
7. Carter KN, Lanumata T, Kruse K, Gorton D. What are the determinants of food insecurity in New Zealand and does this differ for males and females? Aust N Z J Public Health. 2010;34:602-608.
8. Laraia BA, Siega-Riz AM, Gundersen C,

Dole N. Psychosocial factors and socioeconomic indicators are associated with household food insecurity among pregnant women. J Nutr. 2006;136:177-182.

9. Ahmed UI, Ying L, Bashir MK, Abid M, Zulfiqar F. Status and determinants of small farming households' food security and role of market access in enhancing food security in rural Pakistan. PLoS ONE. 2017;12:e0185466.
10. Gregorio MJ, Rodrigues AM, Graca P, de Sousa RD, Dias SS, Branco JC, Canhao H. Food insecurity is associated with low adherence to the Mediterranean diet and adverse health conditions in Portuguese adults. Front Public Health. 2018;6:38.
11. World Health Organization. Adolescent Mental Health: Mapping Actions of Nongovernmental Organizations and Other International Development Organizations. Geneva, Switzerland: World Health Organization; 2012.
12. World Health Organization. 10 Facts on Mental Health, 2014. Available from: http://www.who.int/features/factfiles/mental_health/mental_health_facts/en/ (accessed May 5, 2018).
13. Thomsen PH. Schizophrenia with childhood and adolescent onset—A nationwide register-based study. Acta Psychiatr Scand. 1996;94:187-193.
14. Olweus D. Bully/victim problems among schoolchildren: Basic facts and effects of a school-based intervention program. Eur J Psychol Educ. 1991;17:411-448.
15. Stephens T, Joubert N. The economic burden of mental health problems in Canada. Chronic Dis Can. 2001;22:18-23.
16. United Nations Human Settlements Programme. The Challenge of Slums: Global Report on Human Settlements 2003. London, UK and Sterling, VA, USA: Earthscan Publications Ltd; 2003.
17. Jha DK, Tripathi V. Achieving millennium development goals and India vision 2020: Evidences from the slums of Varanasi city. SAC. 2015;2:4.
18. Census of India. Varanasi Slum Profile at Glance Report, 2011. New Delhi, India: Office of the Registrar General & Census Commissioner, India Ministry of Home Affairs, Government of India; 2011.
19. Coates J, Swindale A, Bilinsky P. Household Food Insecurity Access Scale (hfias) for Measurement of Food Access: Indicator Guide. Washington, DC, USA: Food and Nutrition Technical Assistance Project, Academy for Educational Development; 2007.
20. Kalita BC, Tripathi Y, Garg R, Chaudhary L. Effect of examination stress on cognitive performance and EEG in medical students. Int Arch Integr Med. 2019;6:15-20.
21. Kalita BC, Tripathi Y, Garg R, Chaudhary L. Effect of Examination Stress on Memory and EEG. Ann Int Med Dent Res. 2020;6:6-9.

22. Park K. Park's Textbook of Preventive and Social Medicine. 19th ed. India: Bhanot; 1999. p. 547.
23. Avci D, Selcuk KT, Kaynak S. The magnitude and determinants of emotional-behavioral problems in working adolescents in Turkey. Arch Psychiatr Nurs. 2018;32:44-50.
24. Veit CT, Ware JE. The structure of psychological distress and well-being in general populations. J Consult Clin Psychol. 1983;51:730-742.
25. Bairwa M, Rajput M, Sachdeva S. Modified Kuppuswamy's socioeconomic scale: Social researcher should include updated income criteria. Indian J Community Med. 2013;38:185-186.
26. Agarwal S, Sethi V, Gupta P, Jha M, Agnihotri A, Nord M. Experiential household food insecurity in an urban underserved slum of North India. Food Secur. 2009;1:239-250.
27. Rani D, Singh JK, Acharya D, Paudel R, Lee K, Singh SP. Household Food Insecurity and Mental Health Among Teenage Girls Living in Urban Slums in Varanasi, India: A Cross-Sectional Study. Int J Environ Res Public Health. 2018;15:1-15.
28. Jha DK, Tripathi V. Quality of life in slums of Varanasi city: A comparative study. Transactions. 2014;36:2.
29. Dashiff C, DiMicco W, Myers B, Sheppard K. Poverty and adolescent mental health. J Child Adolesc Psychiatr Nurs. 2009;22:23-32.
30. Lasalvia A, Bonetto C, Tosato S, Zanatta G, Cristofalo D, Salazzari D, Lazzarotto L, Bertani M, Bissoli S, De Santi K, et al. First-contact incidence of psychosis in northeastern Italy: Influence of age, gender, immigration and socioeconomic deprivation. Br J Psychiatry. 2014;205:127-134.
31. Knipe DW, Gunnell D, Pieris R, Priyadarshana C, Weerasinghe M, Pearson M, Jayamanne S, Dawson AH, Mohamed F, Gawarammana I, et al. Is socioeconomic position associated with risk of attempted suicide in rural Sri Lanka? A cross-sectional study of 165,000 individuals. BMJ Open. 2017;7:e014006.
32. Benvegnu LA, Fassa AG, Facchini LA, Wegman DH, Dall'Agnol MM. Work and behavioural problems in children and adolescents. Int J Epidemiol. 2005;34:1417-1424.
33. Sorsdahl K, Slopen N, Siefert K, Seedat S, Stein DJ, Williams DR. Household food insufficiency and mental health in South Africa. J Epidemiol Community Health. 2011;65:426-431.
34. Kessler RC. The effects of stressful life events on depression. Annu Rev Psychol. 1997;48:191-214.
35. Singh J, Singh JV, Srivatava AK, Suryakant. Health status of Adolescent girls in slums of Lucknow. Indian J Community Med. 2006;31:102-103.
36. Reiss F. Socioeconomic inequalities and mental health problems in children and

adolescents: A systematic review. Soc Sci Med. 2013;90:24-31.

37. Kaiser L, Baumrind N, Dumbauld S. Who is food-insecure in California? Findings from the California Women's Health Survey, 2004. Public Health Nutr. 2007;10:574-581.
38. Chilton MM, Rabinowich JR, Woolf NH. Very low food security in the USA is linked with exposure to violence. Public Health Nutr. 2014;17:73-82.
39. Gaur A, Subhash S. Morbidity pattern of adolescents in higher secondary school girls, Rohtak. In: Souvenir XXX National conference Indian Association of Preventive and Social Medicine. Belgaum; 2003. p. 92.
40. Srinivasan K, Prabhu GR. A Study of the Morbidity Status of Children in Social Welfare Hostels in Tirupati Town. Indian J Community Med. 2006;31:3.
41. Singh S, Garg G, Mishra S. A Community Based Study to Know The Impact Of Parental Educational Status On Nutritional Profile Of Adolescent Girls In Rural Areas of Hapur District. Int J Dent Med Sci Res. 2020;4:1-5.
42. Agarwal S, Kumar S, Ingole V, Acharya S, Wanjari A, Bawankule S, Raisinghani N. Does anemia affect cognitive functions in neurologically intact adult patients: a cross-sectional study at rural tertiary care hospital. J Family Med Prim Care. 2019;8:3005-3008.
43. Andrews G, Hall W, Teesson M, Henderson S. The Mental Health of Australians. Canberra, ACT, Australia: Mental Health Branch, Commonwealth Department of Health and Aged Care; 1999.
44. Khedr E, Sherif AH, Elbeih E, Shereef HE, Ahmed Y, Ahmed S. Iron states and cognitive abilities in young adults: neuropsychological and neurophysiological assessment. Eur Arch Psychiatry Clin Neurosci. 2008;258:489-496.
45. International Classification of Disease (ICD-11). Morbidity and Mortality Statistics.
46. Lee A, Lewis M. Testing the Price of Healthy and Current Diets in Remote Aboriginal Communities to Improve Food Security: Development of the Aboriginal and Torres Strait Islander Healthy Diets ASAP (Australian Standardised Affordability and Pricing) Methods. Int J Environ Res Public Health. 2018;15:1585.

https://doi.org/10.46351/jas.v31i2pp115-121

JAS Journal of Anatomical Sciences (U.P. Chapter of Anatomical Society of India)

Original Article

AGE-RELATED CHANGES IN ULTRASONOGRAPHIC SPLEEN LENGTH IN THE ADULT POPULATION OF KANPUR

Alka Singh[1], Hamid Ansari[2], Mohammad Hifzur Rehman[3], Medha Das[2]

1. Department of Anatomy, Autonomous State Medical College, Ayodhya, India
2. Department of Anatomy, Rama Medical College & Research Centre, Kanpur, India
3. Department of Physiology, Career Institute of Medical Sciences & Hospital, Lucknow, India

ABSTRACT

Introduction: The spleen is an intra-abdominal organ affected by a number of diseases. Estimates of spleen size in vivo are often important in the diagnosis, treatment, and prognosis of many disorders which is unreliable by palpation. Several previous studies have sought to develop standards for spleen size such as CT scan, scintigraphy, MRI, and ultrasound. Conventional ultrasound has been shown to be a good measure of spleen size without the need for ionizing radiation. In this study, an attempt was made to determine the normal range of the spleen and its correlation with age of male and female subjects.

Materials and Methods: 80 males and 80 females aged 20 to 60 years from the Department of Anatomy and Radiology, Rama Medical College & Research Centre, Kanpur were selected. Using ultrasound, the length of the spleen was measured.

Results: It was observed that in both men and women, the length of the spleen decreases with age. Spleen length decreases at a slower rate until age 50, after which it declines rapidly. Spleen length is greater in men than in women at all ages.

Conclusions: Spleen size varies greatly among individuals. Establishing normative spleen length data is crucial for assessing changes. Age significantly influences spleen size, especially in males over 50. Consider age before diagnosing spleen length changes.

Keywords : Spleen, Palpation, Hematopoietic system, Portal hypertension

Address for Correspondence:
Dr. Hamid Ansari, Associate Professor, Department of Anatomy, Rama Medical College & Research Centre, Kanpur, India
Email: hamidansari78@gmail.com Mob: 8918748710

Date of Receiving: 13 Oct 2023
Date of Acceptance: 25 Nov 2023
ISSN: 0970-1842

INTRODUCTION

The spleen is an intraperitoneal organ located posterolaterally in the left hypochondrium between the fundus of the stomach and the left hemidiaphragm. In the supine position, the long axis of the spleen is in line with the tenth rib, but in the upright position, it is more vertical. Its extreme or superior angle lies approximately 4 cm from the tenth thoracic spine, and its lateral border at the mid-axillary line in the ninth intercostal space. The shape of the spleen is characteristically tetrahedral but may be modified when enlarged. The splenic hilum is the only portion that is not covered by peritoneum, but here peritoneal reflections carry the main splenic vessels, the splenic arteries, and veins.

The spleen, though not firmly anchored in the body, is attached to the stomach by the gastrosplenic ligament and to the dorsal body wall by the lienorenal ligament. The phrenicocolic ligament, which is not directly attached to the spleen, supports its inferior end. These attachments allow the spleen to enlarge as much as ten times and to shift to ectopic locations. A connective tissue capsule covers the spleen and projects fibers (trabeculae) into its pulp. The peritoneum covers the capsule.

The spleen is an intra-abdominal organ affected by a number of diseases. In various clinical conditions, the spleen is enlarged, most commonly due to reactive proliferation of lymphocytes or reticuloendothelial cells. The spleen also enlarges in malignancies of the hematopoietic system such as lymphoma, diseases associated with portal hypertension, disseminated tuberculosis, malaria, kala azar, cirrhosis, collagen storage diseases, etc.

The normal limits of spleen size must be known for pathological changes to be recognized. It is impractical to calculate the volume of the spleen. For this reason, especially splenic length determination is more meaningful and important [1–3]. Estimates of spleen size in vivo are often crucial in the diagnosis, treatment, and prognosis of many disorders. Accurate measurement of the spleen by palpation is unreliable because in some cases, the spleen may be palpable of normal size while a palpable spleen is not always of normal size. Imaging of the spleen can be obtained with simple radiography [4], but this exposes the patient to avoidable radiation exposure. In addition, if there is an upper lateral quadrant mass, it is very difficult to distinguish splenic tissue from that of nearby organs.

Radionuclide imaging is also used to estimate spleen size. Its accuracy depends on the vascular integrity of the organ and it exposes the patient to excessive gamma radiation [5]. Ultrasound has been shown to be both accurate and reliable for the measurement of splenic dimensions [6]. As a result of recent advances in ultrasonography, assessment of

splenic size (either palpable or unpalpable) has become feasible, reliable, and accurate. Ultrasonography is a simple, safe, and accurate method of assessing splenic size, and patients with persistent splenomegaly should be followed up closely for the development of complications, which may necessitate splenectomy [5].

Ultrasound scanning, apart from being non-ionizing, is painless, non-invasive, widely available, easy to use, and less expensive than most other imaging methods. However, its main limitation is being operator-dependent. It can demonstrate the existence and composition of splenic masses, changes in splenic echotexture and outline, progressive changes in masses, and the size of the spleen.

Measurement of splenic length by ultrasound is reliable within and between technicians. Measurement of splenic width, however, is less reliable, as evidenced by only moderate intra- and inter-rater reliability. This finding supports the historical assessment of splenomegaly based on spleen length.

Because the measurement of splenic width is less reliable, defining splenomegaly on the basis of splenic volume may be more uncertain [7]. In this study, an attempt was made to determine the normal range of the spleen and its correlation with the age of male and female subjects.

MATERIAL AND METHODS

A total of 160 patients, 80 men, and 80 women aged 20 to 60 years were selected for this study. Informed consent was obtained from all patients before inclusion. The study's purpose, potential effects, and examination stages were explained either individually or in groups.

Patients were psychologically reassured and instructed to relax before the examination. Then, they were asked to lie supine on the couch with arms away from the chest wall and to take shallow breaths.

All measurements were conducted on sections through the splenic hilum to ensure a consistent reference point for reproducibility, following the guidelines of the American Institute of Ultrasound in Medicine [8], as described by Lamb et al. [9]. Splenic length (the maximum distance between the dome of the spleen and the splenic tip) was measured on the longitudinal section.

Exclusion criteria: Patients with a history of splenectomy, age under 20, history of malignancy, hematologic disorders, or persistent fever were excluded from the study.

Ethics: Ethical clearance was obtained from the college ethics committee [RMCHRC/Ethics/2022/2035-A].

Age	No. of subject	Mean±SD (cm)	Range (cm)	F ratio
21-30 yrs.	20	10.61 ± 1.35	9.97-11.24	F=9.088
31-40 yrs.	20	10.38 ± 1.81	9.82-10.93	(P<0.001)
41-50 yrs.	20	9.95 ± 1.33	9.32-10.57	
51-60 yrs.	20	8.64 ± 1.33	8.02-9.26	

Table 1. Comparison of splenic length in different age groups in males

Statistical analysis: All data were entered into a Microsoft Excel sheet and then statistically analyzed using SPSS software version 26 (SPSS Inc., Chicago, Illinois, USA). Descriptive statistics including frequency, percentage, mean, and standard deviation were utilized.

RESULTS

Spleen length was 10.61 ± 1.35 cm in the age group of 21-30 years and decreased to 8.64 ± 1.33 cm in the age group of 51-60 years in males [Table-I]. In females, spleen length decreased from 9.63 ± 1.32 cm to 8.41 ± 1.43 cm from the age group of 21-30 years to the age group of 51-60 years [Table-II]. This decrease in length was significant (F=9.088, P<0.001) in males as well as females (F=3.108, P<0.05).

In both males and females, splenic length decreased with age. The splenic length decreased at a slow rate up to the age of 50 years, after which it decreased rapidly. The splenic length was greater in males than females in each age group.

Correlation analysis showed that spleen length was negatively correlated with age in all adults [Table-III]. So, with increasing age, spleen length was found to be decreasing. This decrease in splenic length is significant (Pearson Correlation -0.259, P<0.05) in males and was not significant in females (Pearson Correlation -0.076, P>0.05).

DISCUSSION

The splenic size may provide insights into the diagnosis and prognosis of gastrointestinal and hematologic diseases [10]. Most individuals in this study had spleens less than 11 cm in length, consistent with Frank et al.'s findings [5].

According to Rosenberg et al. [11], girls aged 15 years or older have spleens with an upper normal limit of 12 cm, slightly differing from this study's results. In this study, splenic length gradually declined until age 50, then reduced rapidly, consistent with Loftus and Matrewali's findings [12]. They noted a significant increase in splenic length up to age 20, followed by a moderate decline in growth rate until age 50.

Age	No. of subject	Mean±SD (cm)	Range (cm)	F ratio
21-30 yrs.	20	9.63 ± 1.32	9.01-10.25	F=3.108
31-40 yrs.	20	9.41 ± 1.27	8.81-10.00	(P<0.05)
41-50 yrs.	20	9.21 ± 1.34	8.57-9.84	
51-60 yrs.	20	8.41 ± 1.43	7.74-9.08	

Table 2. Comparison of splenic length in different age groups in females

Konus et al. [13] found the best correlation between splenic length and body height. Splenic dimensions (length, breadth, and thickness) decreased with age in both males and females. Age showed a statistically significant negative correlation with splenic measurements in males [14].

Similarly, research on Chinese individuals showed rapid splenic length increase up to age 20, followed by gradual decline until age 50, then rapid decrease. The average spleen measured 9.56 ± 1.37 cm, with a negative correlation between age and splenic length [15].

Studies in Tripura, West Nepal, East Nepal, and North India found an inverse relationship between adult subjects' age and splenic size, indicating a decrease with age [16]. Ezeofor et al. [17] studied children aged 5 to 17 years, noting a significant correlation between splenic length and age ($P < 0.001$), with males having statistically longer spleens than females.

CONCLUSION

Spleen size varies greatly among individuals, emphasizing the need for normative data to assess changes accurately. Patient age

Parameters	Splenic length(cm)			
	Male (80)		Female (80)	
	Pearson's Correlation	P value	Pearson's Correlation	P value
Age(years)	-0.259	<0.05	-0.076	>0.05

Table 3. Correlation of spleen length and age

significantly influences spleen size. Therefore, age consideration is crucial when evaluating spleen length changes due to disease. In males over 50, spleen size is typically reduced, warranting caution in reporting normal spleen size.

REFERENCES

1. Megremis SD, Vlachonikolis IG, Tsilimigaki AM. Spleen length in childhood with US: normal values based on age, sex, and somatometric parameters. Radiology. 2004 Apr;231(1):129-34. doi: 10.1148/radiol.2311020963.
2. Safak AA, Simsek E, Bahcebasi T. Sonographic assessment of the normal limits and percentile curves of liver, spleen, and kidney dimensions in healthy school-aged children. J Ultrasound Med. 2005 Oct;24(10):1359-64. doi: 10.7863/jum.2005.24.10.1359.
3. Watanabe Y, Todani T, Noda T, Yamamoto S. Standard splenic volume in children and young adults measured from CT images. Surg Today. 1997;27(8):726-8. doi: 10.1007/BF02384985.
4. Schindler G, Longin F, Helmschrott M. [The individual limit of normal spleen size in routine x-ray film (author's transl)]. Radiol. 1976 Apr;16(4):166-71.
5. Frank K, Linhart P, Kortsik C, Wohlenberg H. [Sonographic determination of spleen size: normal dimensions in adults with a healthy spleen]. Ultraschall Med. 1986 Jun;7(3):134-7. doi: 10.1055/s-2007-1011931.
6. Petzoldt R, Lutz H, Ehler R, Neidhardt B. [Determination of splenic size by ultrasonic scanning (author's transl)]. Med Klin. 1976 Nov;71(48):2113-6.
7. Hosey RG, Mattacola CG, Kriss V, Armsey T, Quarles JD, Jagger J. Ultrasound assessment of spleen size in collegiate athletes. Br J Sports Med. 2006 Mar;40(3):251-4; discussion 251-4. doi: 10.1136/bjsm.2005.022376.
8. Al-Salem AH, Al-Aithan S, Bhamidipati P, Al-Jam'a A, Al Dabbous I. Sonographic assessment of spleen size in Saudi patients with sickle cell disease. Ann Saudi Med. 1998;18(3):217-20. doi: 10.5144/0256-4947.1998.217.
9. Lamb PM, Lund A, Kanagasabay RR, Martin A, Webb JA, Reznek RH. Spleen size: how well do linear ultrasound measurements correlate with three-dimensional CT volume assessments? Br J Radiol. 2002 Jul;75(895):573-7. doi: 10.1259/bjr.75.895.750573.
10. Niederau C, Sonnenberg A, Müller JE, Erckenbrecht JF, Scholten T, Fritsch WP. Sonographic measurements of the normal liver, spleen, pancreas, and portal vein. Radiology. 1983 Nov;149(2):537-40. doi: 10.1148/radiology.149.2.6622701.
11. Rosenberg HK, Markowitz RI, Kolberg H, Park C, Hubbard A, Bellah RD. Normal splenic size in infants and children: sonographic measurements. AJR Am J Roentgenol. 1991 Jul;157(1):119-21. doi: 10.2214/ajr.157.1.2048509.

12. Loftus WK, Metreweli C. Normal splenic size in a Chinese population. J Ultrasound Med. 1997 May;16(5):345-7. doi: 10.7863/jum.1997.16.5.345.
13. Konuş OL, Ozdemir A, Akkaya A, Erbaş G, Celik H, Işik S. Normal liver, spleen, and kidney dimensions in neonates, infants, and children: evaluation with sonography. AJR Am J Roentgenol. 1998 Dec;171(6):1693-8. doi: 10.2214/ajr.171.6.9843315.
14. Chime PE, Oti B, Okenwa WO. The Relationship between Splenic Length and Age in Adult Nigerians. J Biosci Med. 2020;8(4):121-7. doi: 10.4236/jbm.2020.84011.
15. Chakraborti S, Saha N, Debbarma B, Das S, Leishram D. Normal Spleen Length by Ultrasonography in Adults of Tripura. 2016 Feb;15:55-60.
16. Ezeofor SN, Obikili EN, Anyanwu GE, Onuh AC, Mgbor SO. Sonographic assessment of the normal limits of the spleen in healthy school children in South-East Nigeria. Niger J Clin Pract. 2014;17(4):484-8. doi: 10.4103/1119-3077.134046.
17. Mohtasib RS, et al. Sonographic measurements for spleen size in healthy Saudi children and correlation with body parameters. Ann Saudi Med. 2021;41(1):14-23. doi: 10.5144/0256-4947.2021.14.

https://doi.org/10.46351/jas.v31i2pp122-131

JAS Journal of Anatomical Sciences (U.P. Chapter of Anatomical Society of India)

Original Article

MORPHOMETRY OF PROXIMAL FEMUR AND CORRELATIONS OF THE HEAD NECK SHAFT ASSEMBLAGE

Ajay Verma[1], Garima Sehgal[1], Mariam Moonis[1], Navneet Kumar[1]

1. Department of Anatomy, King George's Medical University, Lucknow, India

ABSTRACT

Introduction: Recent times have shown an increased incidence of proximal femur pathologies. The morphology and morphometry of the proximal femur is an important component of hip biomechanics. Altered diameters and angles affect the range of motion and have been found to be causative in hip pathologies. Understanding femoral shape and orientation can help to predict degenerative diseases, relate to hip instability, in designing hip replacement implants, in preoperative planning and for forensic identification of human remains. This study aimed at determining the morphometric parameters of proximal femur and to analyse the correlations between measured parameters.

Materials and Methods: Measurements were made on 120 dry adult human femora of unknown sex (Right-60, Left-60). Transverse and vertical diameters of the head and neck and anterior and posterior neck lengths were measured using digital vernier calliper and neck-shaft angle was measured using a goniometer. Measurements were statistically analysed. Correlation coefficient was used to analyse the relationship among variables.

Results: Mean vertical and transverse diameters of head were 43.23 mm and 43.16 mm and Mean Anterior and Posterior neck lengths were 28.37 mm and 35.81 mm respectively. Mean Neck shaft angle was 128.82□. On comparison between sides, statistically significant differences were observed in neck shaft angle and anterior neck length measurements. Correlation between neck shaft angle and neck length as well as between neck shaft angle and head diameters revealed variable patterns on left and right sides.

Conclusions: Our study has found a significant difference in anterior length of neck in left and right femur as well as a substantial and statistically significant positive correlation was observed between anterior neck length and neck shaft angle of the right femur.

Keywords : Femur, Morphometry, Vernier, Goniometer, Hip, Prosthesis

Address for Correspondence:
Dr. Garima Sehgal, Professor (Jr. Grade), Department of Anatomy, King George's Medical University, Lucknow, India
Email: garimasehgal@kgmcindia.edu Mob: 9044513758

Date of Receiving: 06 Nov 2023
Date of Acceptance: 09 Dec 2023
ISSN: 0970-1842

INTRODUCTION

Femur and tibia form the greatest proportion of body stature and are of great importance for anthropologists and forensic experts. Various studies and methods have derived equations for femur morphometry, aiding in the estimation of stature across different races, age groups, genders, and populations, as well as assisting forensic experts. Orthopedic surgeons encounter various pathological conditions, congenital anomalies, and fractures related to the femur, and treatment can be planned using knowledge of morphometry [1].

The length of the femur is about one-fourth of the height of an individual. It consists of three parts: the upper end (proximal end), shaft, and lower end (distal end). The upper end comprises the head, neck, greater trochanter, and lesser trochanter. The head is often described as more than half a sphere, with a small rough fovea present postero-inferiorly, protruding medially from its short neck. The intracapsular head is encircled by the acetabular labrum distally to articulate with the acetabulum. The femoral neck has rounded contours with a slightly concave upper surface and an obliquely straight lower surface.

It is approximately 5 cm long, narrowest in its midpart, widest at the sides, and connects the head to the shaft at an average angle of 127°. The neck-shaft angle facilitates movement at the hip joint and provides leverage for the muscles acting around the hip joint, which are attached to the proximal femur. The neck-shaft angle is widest at birth and gradually decreases until age 10. The neck is laterally rotated to the shaft (angle of anteversion) by about 10–15°, although values of this angle vary between individuals and populations [2].

The incidence of femur fractures is higher in road traffic accidents, particularly among the geriatric population. Studies have also reported an increased rate of proximal femur fractures, intertrochanteric femur fractures, hip osteoarthritis, with most ailments being treated surgically using prostheses. The implants and prostheses available are designed based on European data [3]. The morphometry of the femur varies with age, race, gender, etc., so this should be considered when designing suitable implants [4].

We were unable to find enough literature on morphometric studies of the proximal femur among the population of Uttar Pradesh. Hence, this study was planned to address this gap. The primary objectives of the study were (i) to measure the indices of the proximal femur and (ii) to observe correlations between various measured bony parameters.

MATERIAL AND METHODS

The study was conducted in the Department of Anatomy, KGMU, UP. Observations were

made on the study sample which included 120 dry adult human femora of unknown sex (Right-60, Left-60), procured from the Bone Bank of the Department of Anatomy, King George's Medical University, Lucknow, UP. All measurements were taken using a Digital vernier caliper and Goniometer. The bones with visible deformities, fractures, and the bones that were broken were excluded from the study. Observations were made with respect to the following parameters:

1. Vertical and Transverse diameters of the head were measured using a digital vernier caliper as the distance between the most superior and inferior points on the articular margin of the head in the vertical plane (Fig. 1A) and the maximum distance of the femoral head on the articular margin of the head in the horizontal plane respectively (Fig. 1B).

2. Anterior and Posterior neck lengths were measured using a digital vernier caliper as the distance between the base of the head and the midpoint of the intertrochanteric line anteriorly (Fig. 2A) and the distance between the base of the head and the midpoint of the intertrochanteric crest posteriorly respectively (Fig. 2B).

3. Neck-shaft angle was measured using a goniometer as the angle between the long axis of the shaft of the femur and the axis of the neck of the femur (Fig. 3).

All measurements were performed twice by a single person, and then the arithmetic average was taken. The data were tabulated on a Microsoft Excel sheet.

Statistical analysis

All data were statistically analyzed as per standard statistical methods. Variables were compared using appropriate statistical tests to analyze significant effects. A P-value of less than 0.05 was considered statistically significant. The correlation coefficient was used to analyze the relationship among variables.

RESULTS

Mean Vertical diameter of the head was 43.23±3.26 mm. Mean Transverse diameter of the head was 43.16±3.26 mm. The mean anterior neck length was 28.37±3.32 mm. Mean Posterior neck length was 35.81±3.64 mm. Mean Neck shaft angle was 128.82±6.82 (Table 1).

On comparing the measurements of Vertical and Transverse diameters of the head between the right and left sides, there was no statistically significant difference (p-values > 0.05). The averages and standard deviations for these measurements were quite similar for both sides (Table 2). The mean anterior neck length for the left femur was 29.29 mm, and that of the right femur was 27.46 mm.

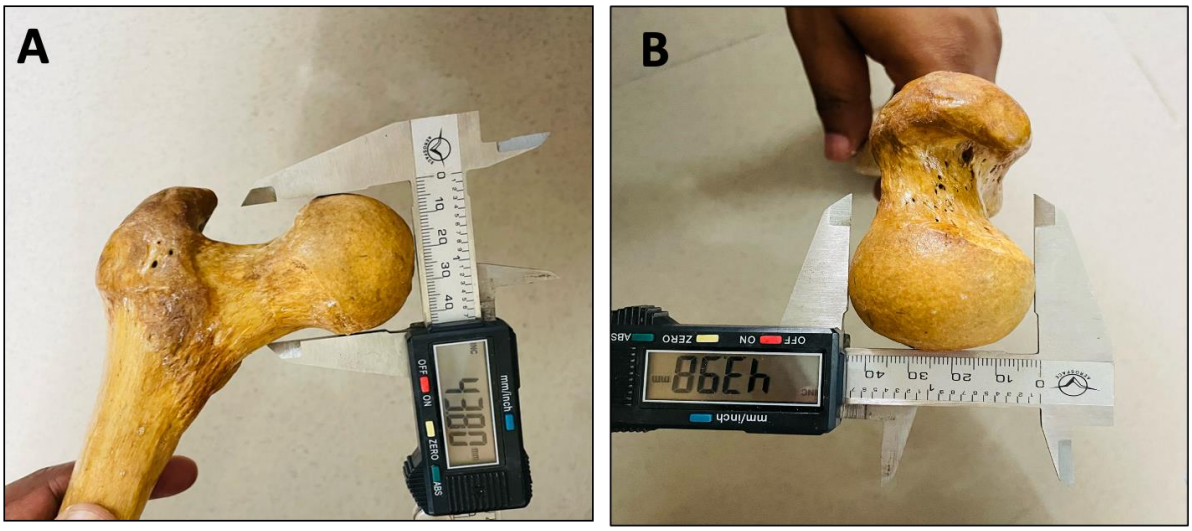


Fig. 1. Measurement of vertical diameter (A) and transverse diameter (B) of head of Femur using digital vernier caliper

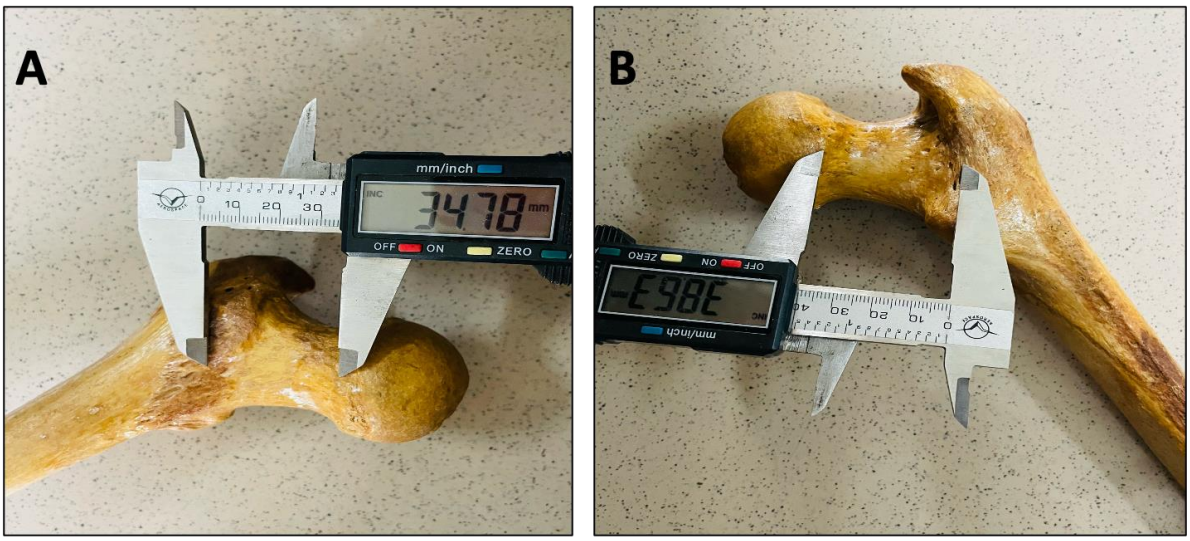


Fig. 2. Measurement of anterior neck length (A) and posterior neck length (B) of head of Femur using digital vernier caliper

On comparing the anterior neck length measurement for the femur of the right and left sides, the difference was statistically significant (p-value = 0.002) (Table 2). The mean posterior neck length measured 35.64 mm on the left side and 35.99 mm on the right side, and the difference was not found to be statistically significant (p-value = 0.596) (Table 2). The mean neck shaft angle was greater on the right side compared to the left side, and a highly significant difference was observed between the left and right femurs (p-value < 0.001) (Table 2).

On analysis of the correlation between neck shaft angle and other parameters of the left

Fig. 3. Measurement of neck-shaft angle of Femur using Goniometer

femur; a weak positive correlation was found between neck shaft angle and the vertical and transverse diameters of the head, and a weak negative correlation was found between neck shaft angle and the anterior and posterior neck lengths. No strong or statistically significant correlation was observed between neck shaft angle and the other measured variables on the left femur (Table 3).

On the analysis of the correlation between neck shaft angle and other parameters of the right femur, a variable and different pattern of correlation was observed compared to the left side. A slight positive correlation was observed between neck shaft angle and the vertical diameter of the head; a weak positive correlation was observed between neck shaft angle and the transverse diameter of the head, and the correlation of head diameters was not found to be statistically significant. A weak positive correlation was observed between neck shaft angle and posterior neck length, which was not significant (Table 4).

A substantial and statistically significant positive correlation was observed between neck shaft angle and anterior neck length ($p = 0.005$), suggesting that as the neck shaft angle increases, there tends to be an increase in anterior neck length of the right femur (Table 4).

DISCUSSION

The cases of total hip arthroplasties are on the increase worldwide, including in India. In this respect, the morphometric measurements of the proximal femur become important as they help in the design of better-fit implants and prostheses for better surgical outcomes. The design and accurate assessment of head prosthesis are major components of arthroplasty. Mismatch of the same may predispose to dislocation, imperfect biomechanics, and groin pain [5].

	Mean	SD	Median	Min	Max	Percentile 25	Percentile 75	Valid N
VERTICAL DIAMETER OF HEAD	43.23	3.26	43.59	28.84	50.75	41.42	45.27	120
TRANSVERSE DIAMETER OF HEAD	43.16	3.26	43.52	29.15	50.74	41.24	44.96	120
ANTERIOR NECK LENGTH	28.37	3.32	28.42	19.74	38.52	25.74	30.38	120
POSTERIOR NECK LENGTH	35.81	3.64	35.77	23.55	44.98	33.56	38.48	120
NECK SHAFT ANGLE	128.82	6.82	130.00	110.00	140.00	122.00	135.00	120

Table 1. Descriptive statistics of all measurements of the Femur (n=120)

In our study, the mean vertical diameter of the head was 43.23±3.26, which was concordant with the study done in the Chinese population where the vertical diameter of the head was 44.64±3.13 mm [6]. Another study on 50 femurs belonging to the Bihar population reported the mean vertical diameter of the head as 40.97±3.46 mm [7]. Dwivedi AK et al [8] conducted a study in the Maharashtrian population and reported the mean vertical diameter of the head as 40.53±3.51mm. Jaiswal et al [9] reported the values as 33.23±4.08 mm.

In our study, the mean transverse diameter of the head was 43.23±3.26 mm, which was higher than studies done by Sinha et al and Dwivedi et al [7,8]. Sinha SK et al studied 50 femurs of the population of Bihar and reported a mean transverse diameter of the head of 41.74±2.76 mm. Dwivedi AK et al [8] conducted a study in the Maharashtrian population and reported the mean transverse diameter of the head as 40.44±3.47 mm. Katch AU et al [6] conducted a study in the Chinese population and reported the values as 44.55±3.37 mm, which were higher than our values.

Optimum neck morphometrics allow the femur to adjust to the biomechanics of the implant. The mean anterior neck length in our study was 28.37±3.32 mm. Sinha SK et al [7] studied 50 femurs of the population of Bihar and reported that the mean anterior neck length was 29.75±5.30 mm. Dwivedi AK et al [8] conducted a study in the Maharashtrian population and reported the mean anterior neck length as 29.92±4.04 mm. Katchy AU et al [6] conducted a study in the Chinese population and reported the values as 31.86±4.22 mm.

The mean posterior neck length in our study was 35.81±3.64 mm, which was similar to Sinha et al and Dwivedi et al. Sinha SK et al [7,8] studied 50 femurs from Bihar and reported values as 35.03±4.87 mm. Dwivedi AK et al [8] conducted a study in the Maharashtra and reported the mean posterior neck length as 35.23±4.22 mm. Jaiswal et al [9] reported the values as 35.59±3.74 mm.

		Mean	SD	Median	Min	Max	Percentile 25	Percentile 75	Valid N
LEFT FEMUR	VERTICAL DIAMETER OF HEAD	42.90	3.83	43.62	28.84	50.75	40.57	45.41	60
	TRANSVERSE DIAMETER OF HEAD	42.75	3.78	43.50	29.15	50.74	40.59	44.91	60
	ANTERIOR NECK LENGTH	29.29	3.38	29.28	19.74	38.52	27.40	31.13	60
	POSTERIOR NECK LENGTH	35.64	4.05	35.77	23.55	43.93	33.40	38.58	60
	NECK SHAFT ANGLE	124.35	5.72	125.00	110.00	140.00	120.00	130.00	60
RIGHT FEMUR	VERTICAL DIAMETER OF HEAD	43.56	2.57	43.58	37.85	49.71	42.04	45.25	60
	TRANSVERSE DIAMETER OF HEAD	43.56	2.62	43.55	37.72	50.59	42.32	44.96	60
	ANTERIOR NECK LENGTH	27.46	3.01	27.63	20.22	36.33	25.23	29.58	60
	POSTERIOR NECK LENGTH	35.99	3.21	35.84	28.26	44.98	33.58	38.25	60
	NECK SHAFT ANGLE	133.28	4.54	133.50	120.00	140.00	130.00	135.00	60

Table 2. Comparison of measurements of various parameters of Femur between left and right

The neck shaft angle (NSA), which helps the acetabulum align with the femoral head, is of great structural and diagnostic value in hip joint mechanics. The angle is a beneficial structural adaptation that increases hip rotation and helps the lower limb to swing away from the pelvis, increasing freedom of movement [10]. In our study, the neck shaft angle was 128.82±6.82. The values are in the range of reported literature. Sinha SK et al [7] reported the mean neck shaft angle as 125.96±6.10. Katchy AU et al [6] reported the mean neck shaft angle as 132.15±7.30. These values were higher as the sample size was 716, and the population group was Chinese, which was different from our study.

Aparna et al [11] reported the mean neck shaft angle as 125 degrees. Ravi et al [1] in their study reported the neck shaft angle as 136.8±4.45. Kamath et al [12] reported in their study the mean neck shaft angle as 137.80±6.90. Kulkarni et al [4] performed a CT-based measurement of the head of the femur, and the values were 127.2±5.2.

K. Sreenivasa Reddy et al [13] conducted a study in the south Indian population and reported the neck shaft angle as 125.35±7.883. The neck shaft angle of the standard femoral prosthesis for arthroplasty is 131°. This is greater than the mean neck shaft angle observed in our study.

	NECK SHAFT ANGLE	ANTERIOR NECK LENGTH	POSTERIOR NECK LENGTH	VERTICAL DIAMETER OF HEAD	TRANSVERSE DIAMETER OF HEAD
Spearman's rho Correlation Coefficient	1.000	-.058	-.191	.050	.067
Sig. (2-tailed)	.	.661	.143	.703	.613
N	60	60	60	60	60

Table 3. Correlation analysis between neck-shaft angle and various other parameters of left Femur

An inappropriate neck shaft angle may increase difficulty in the surgical procedure during arthroplasty and can later impair the natural biomechanics of the hip joint during recovery [14]. In the case of angle mismatch in implants, there could be deformities such as valgus in high angle implants and varus deformities in cases of low angle implants leading to alterations in the biomechanics of the hip and knee joints [13]. This information may thus be useful in guiding prosthetists and orthopedic surgeons to construct suitable implants. The present study reports a significant difference between NSA of the right and left femurs (p-value < 0.001). This suggests a substantial anatomical distinction between the two sides, particularly in terms of the neck shaft angle. In our study, a notable and statistically significant positive correlation was found between the neck shaft angle and the anterior neck length of the right femur.

It can be observed that values for various bony parameters at the upper end of the femur display regional variations in India. Anthropometric measurements for the normal upper end of femora are also variable among western and Indian populations. The environment plays an important role in development; the regional variation of the femur bone is influenced by the geographical area, sex, stature, and heredity. As the proximal end of the femur varies in different ethnic groups with respect to their build, physique, habits, and genetic makeup, the anthropometric measurements of the proximal femur can serve as a valuable tool for designing better-fitting and well-adjusted femoral implants and prostheses to improve treatment outcomes.

	NECK SHAFT ANGLE	ANTERIOR NECK LENGTH	POSTERIOR NECK LENGTH	VERTICAL DIAMETER OF HEAD	TRANSVERSE DIAMETER OF HEAD
Spearman's rho Correlation Coefficient	1.000	.355**	.119	.153	.141
p-value	.	.005	.365	.244	.283
N	60	60	60	60	60

Table 3. Correlation analysis between neck-shaft angle and various other parameters of right Femur

Limitations of the study: The study was done on 120 femora and multiple parameters were analyzed. A variable pattern of correlations were obtained between parameters on left and right side and a strong and statistically significant correlation was observed between neck shaft angle and anterior length of neck on the right side.

The sample size can be expanded further and studied to confirm and further studies can be planned in future to assess the morphometry of distal end and correlation for providing further insight.

CONCLUSION

The knowledge of anatomy of proximal end of femur is very important for clinical and pathological states of the hip joint and for designing prostheses for hip replacement. Previous literatures have reported femoral morphometry to vary with age, gender, race, ethnicity etc. Regional differences among different groups of populations exist.

Though these differences have been explored much in other parts of the world, fewer studies have been conducted among the Indian population comprising of a heterogenous population of varying morphological subgroups. Our study attempted to bridge this gap and offers to give valuable insights for further exploration. A notable and statistically significant positive correlation was found between neck shaft angle and anterior neck length of right femur.

REFERENCES

1. Ravi GO, Shaik Hussain Saheb, Abraham Ratna Joseph N. A Morphometric Study of Femur and Its Clinical Importance. Int J Integr Med Sci. 2016;3(7):341-44.
2. Gray's Anatomy. 41st ed. Churchill Livingstone: Elsevier; 2016. p. 1348-1350.
3. Jadawala V, Deshpande S, Taywade S, et al. A Review on Morphometry of the proximal femur and its clinical significance. J Pharm Negative Results. 2022;13(9):2855-2863.
4. Kulkarni M, Naik AM, Shetty CB, Paruthikunnan SM, Rao SK. CT based measurement of anatomical dimensions of femur and its relevance in nail designs for proximal femoral fractures. J Orthop. 2019 Dec 10;20:63-69.
5. Girard J. Femoral head diameter considerations for primary total hip arthroplasty. Orthop Traumatol Surg Res. 2015 Feb;101(1 Suppl):S25-9.
6. Katchy AU, Nto NJ, Agu AU, Ikele IT, Chime SC, Ugwu AU. Proximal femoral geometry analysis of Igbos of South East Nigeria and its clinical application in total hip replacement and hip surgeries: A dry bone study. Niger J Clin Pract. 2021;24:369-79.
7. Sinha SK, Suman S, Prasad S, Hayat SMB. Study of Dimensions of Head and Neck of Human Femur and its Clinical Significance. Int J Anat Res. 2022;10(2):8359-8362.
8. Dwivedi AK, Airan N, Bhatnagar R. Estimation of Length of Femur from its

Proximal Segment in Maharashtrian Population. 2019;8(1):AO01-AO05.

9. Jaiswal P, Verma L, Ghulyani T. Correlation between femoral offset and other morphometric parameters. Natl J Clin Anat. 2017;6(2):137-145.
10. Isaac B, Vettivel S, Prasad R, Jeyaseelan L, Chandi GP. Prediction of the femoral neck-shaft angle from the length of the femoral neck. Clin Anat. 1997;10:318–323.
11. Gullapalli A, Inuganti AK. Morphometric study of femoral neck-shaft angle and its clinical significance. Int J Anat Res. 2017;5(3.2):4261-64.
12. Kamath SU, Agarwal S, Austine J. Morphology of Proximal Femur in South-West Coast of India. Malays Orthop J. 2020;14(3):143-150.
13. Reddy KS, Kumar PDA. Morphometry of proximal end of femur in population of Telangana state and its clinical application. Indian J Clin Anat Physiol. 2019;6(1):57-60.
14. Gupta M, Devadas D, Sahni C, et al. Morphometric Analysis of the Proximal Femur With Its Clinical Correlation in Eastern Uttar Pradesh Region. Cureus. 2022;14(9):e28780.

https://doi.org/10.46351/jas.v31i2pp132-140

JAS Journal of Anatomical Sciences (U.P. Chapter of Anatomical Society of India)

Original Article

CLINICAL SIGNIFICANCE OF MORPHOLOGICAL AND MORPHOMETRICAL ANALYSIS OF FORAMEN MAGNUM

Sushma Mishra[1], Prerna Gupta[1], Hari Prasad[1]

1. Department of Anatomy, T.S. Mishra Medical College & Hospital, T.S. Mishra University, Anora, Amausi, Lucknow, India

ABSTRACT

Introduction: The foramen magnum is a transitional zone between cranial cavity and spinal canal, and it is related with the very important neuro-vascular structures like vertebral arteries, spinal accessory nerves and spinal arteries and terminal part of medulla oblongata. Therefore, thorough knowledge of morphometric and morphological variation of foramen magnum is important for authentic radiological diagnosis and surgeries in the area of cranio-cervical junction.

Materials and Methods: This study was done on 70 dried human skulls taken from the Department of Anatomy, TS Misra Medical College & Hospital, Era's Lucknow Medical College & Hospital, and KGMU, Lucknow. The shape of foramen magnum was observed and transverse and antero-posterior (AP) diameters of foramen magnum (FM) were measured with the help of vernier caliper. Foramen magnum index (FMI) and foramen magnum area (FMA) were also calculated.

Results: The study showed six types of shapes of foramen magnum. The oval shaped foramen magnum was found in 47% (in 33 skulls), round 30% (in 30 skulls), tetragonal 10 % (in 10 skulls), triangular & irregular 10% (in 5-5 skulls), pentagonal 2% (in 2 skulls), and hexagonal 1% (in single skull). The mean anteroposterior and transverse diameter were 34.3±2.90mm and 28.9±2.80 mm respectively. The mean foramen magnum index and Foramen magnum area were 77.80±27.80 and 845.90±87.50 mm^2 respectively.

Conclusions: The data obtained from the present study will be useful to the neurosurgeons prior to the surgeries in the area of cranio-cervical junction and posterior cranial fossa. The morphometric and morphological knowledge of the foramen Magnum has important clinical implications in the prognosis and treatment of various neurological pathologies like Arnold Chiari syndrome, Achondroplasia and posterior cranial fossa lesions.

Keywords : Foramen magnum, Transcondylar approach, Foramen magnum index, Foramen magnum area, Transverse diameter (TD), Anteroposterior diameter, Trigonal.

Address for Correspondence:
Dr. Sushma Mishra, Tutor, Department of Anatomy, T.S. Mishra Medical College & Hospital, T.S. Mishra University, Anora, Amausi, Lucknow, India
Email: sushma27mishra@gmail.com Mob: 9044513758

Date of Receiving: 18 Nov 2023
Date of Acceptance: 25 Dec 2023
ISSN: 0970-1842

INTRODUCTION

The foramen magnum is an intraosseous foramen located at the anteromedian position in the occipital bone of the skull [1]. It provides a passageway to the lower end of the medulla oblongata with its meninges, vertebral arteries and veins, spinal accessory nerves, spinal arteries, apical ligament of the dens, and the tectorial membrane [1]. Knowledge of anatomical variations of the foramen magnum is important in terms of its morphology as well as morphometry because in cases of congenital abnormalities and pathologies at the cranio-cervical junction, the morphometry and morphology of the foramen magnum are greatly affected.

Therefore, neurosurgeons should be aware of the anatomical variations of the foramen magnum and its related structures prior to surgery. Additionally, knowledge of anatomical variation of the foramen magnum also helps physical anthropologists and forensic experts in the identification of mutilated bodies in conditions of warfare, nuclear explosions, or natural disasters.

Very little work has been done on the basic morphometric and morphological variations of the foramen magnum [2]. To the best of our knowledge, to date, there is only one research study done by Philipp Guber et al., where they attempted to explore any changes in biological characters of the foramen magnum that occur from generation to generation [2].

The aim of the present study was to determine and analyze the morphometric and morphological variations of the foramen magnum, which will aid neurosurgeons prior to surgeries conducted at the cranio-cervical junction in cases of cerebellar herniation or achondroplasia.

MATERIAL AND METHODS

This observational study was carried out on 70 dried adult human skulls of unknown age and gender. The skulls were obtained from the Department of Anatomy, TS Misra Medical College, Era's Lucknow Medical College & Hospital, and KGMU, Lucknow. Approval from the Institutional Ethics Committee was obtained prior to the study. Fully cleaned, undamaged skulls were selected, whereas deformed and damaged skulls were excluded. The shape of the foramen magnum was observed. The anteroposterior and transverse diameters were measured with the help of a digital vernier caliper. The precision of the caliper's measurement was 0.01mm. The FMI (foramen magnum index) and FMA (Foramen magnum Area) were also calculated and registered in tabulated form.

The anteroposterior diameter (AP) was measured from basion (the midpoint of the anterior margin of the foramen magnum) to opisthion (the midpoint of the posterior margin of the foramen magnum). The transverse diameter (TD) was measured between the

maximum concavity of the right and left lateral margins of the foramen magnum (Fig.1).

The Foramen magnum Index (FMI) and Foramen magnum area (FMA) were calculated by the following formulae:

FMI = (Transverse diameter × 100) / Anteroposterior diameter [3]

Radinsky formula: FMA = ¼ × π × TD × AP^3

Where "π" was accepted as 3.14 in both formulas.

Statistical Analysis:

All the data was expressed as mean ± SD and all the statistical analysis were done by using the SPSS software version16.0.

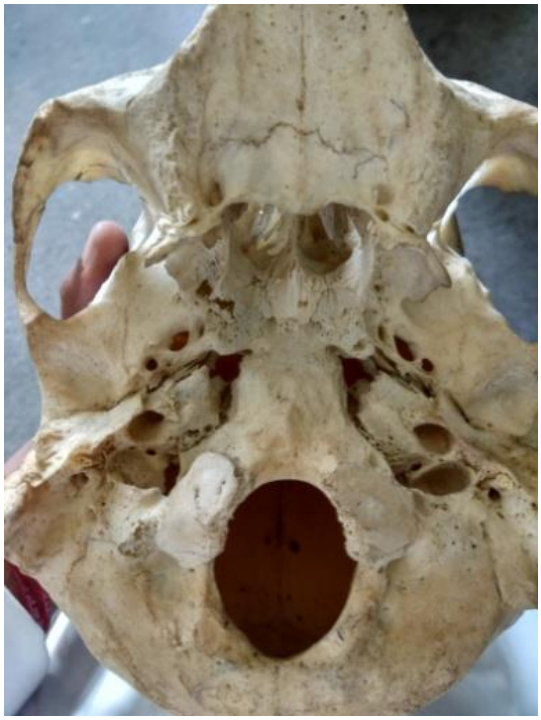


Fig. 1. Land marks and dimensions of foramen magnum

RESULTS

In the current study, the morphometric and morphological parameters were studied in 70 dried adult human skulls of unknown age and gender from the North Indian population. The most common shape was an oval-shaped foramen magnum found in the maximum number of skulls (33 skulls), accounting for 47.1% of the total.

The second most common shape was a round-shaped foramen magnum, found in 16 skulls, representing 22.8%. Tetragonal-shaped foramen magnum was found in 10 skulls, accounting for 14.2%, irregular-shaped foramen magnum in 5 skulls (7.1%), and hexagonal-shaped foramen magnum in a single skull (1.7%).

S. No.	Shapes of FM	Number of FM	Percentage %
1	Oval	33	47.1%
2	Round	16	22.8%
3	Trigonal	5	7.1%
4	Tetragonal	10	14.2%
5	Hexagonal	1	1.7%
6	Irregular	5	7.1%

Table 1. Number of different shapes of foramen magnum and their percentage

A new variant, "trigonal" shaped foramen magnum, was found in 5 skulls (7.1%). Table 1 and Figure 2 illustrate the different shapes of the foramen magnum observed in the current study. Table 2 shows all the morphometric and morphological parameters of the foramen magnum according to the different shapes found in this study.

The mean anteroposterior (AP) and transverse diameters (TD) of oval-shaped foramen magnum were 35.0±3.1 mm and 28.7±2.2 mm, respectively, with mean FMA and FMI values of 826.7±84.0 mm^2 and 78.8±28.0, respectively. For round-shaped foramen magnum, the mean AP and TD were 33.5±2.9 mm and 30.9±3.1 mm, with mean FMA and FMI values of 951.6±39.0 mm^2 and 81.2±28.4, respectively.

Trigonal-shaped foramen magnum had mean AP, TD, FMA, and FMI values of 34.6±1.6 mm, 27.6±1.9 mm, 764.7±25.0 mm^2, and 74.9±27.3, respectively. Tetragonal-shaped foramen magnum had mean AP, TD, FMA, and FMI values of 32.4±2.5 mm, 29.0±3.2 mm, 868.2±77.0 mm^2, and 73.8±27.1, respectively.

Hexagonal-shaped foramen magnum was observed in one skull (1.7%), with AP and TD diameters of 35.0±0.0 mm and 26.0±1.0 mm, respectively. The foramen magnum index for hexagonal-shaped foramen magnum was 70.6±26.5, and the foramen magnum area was 778.7±43.0 mm^2. Irregular-shaped foramen magnum had mean AP, TD, FMA, and FMI values of 34.6±1.8 mm, 26.0±1.0 mm, 778.7±43.0 mm^2, and 70.6±26.5, respectively. Table 3 shows the morphometric parameters of the total studied skulls (70 skulls). The mean AP and TD dimensions of the foramen magnum of all 70 skulls were 34.3±2.90 mm and 28.9±2.80 mm, respectively, with mean FMI and FMA values of 77.80±27.80 and 845.90±87.50 mm^2.

The minimum mean values of AP and TD of the foramen magnum for all skulls were 32.44 mm and 24.00 mm, respectively, while the minimum FMI and FMA values were 73.80 and 611.72 mm^2. The maximum mean values of AP and TD of the foramen magnum for all studied skulls were 35.09 mm and 30.90 mm, respectively, with the maximum mean values of FMI and FMA being 88.05 and 851.93 mm^2.

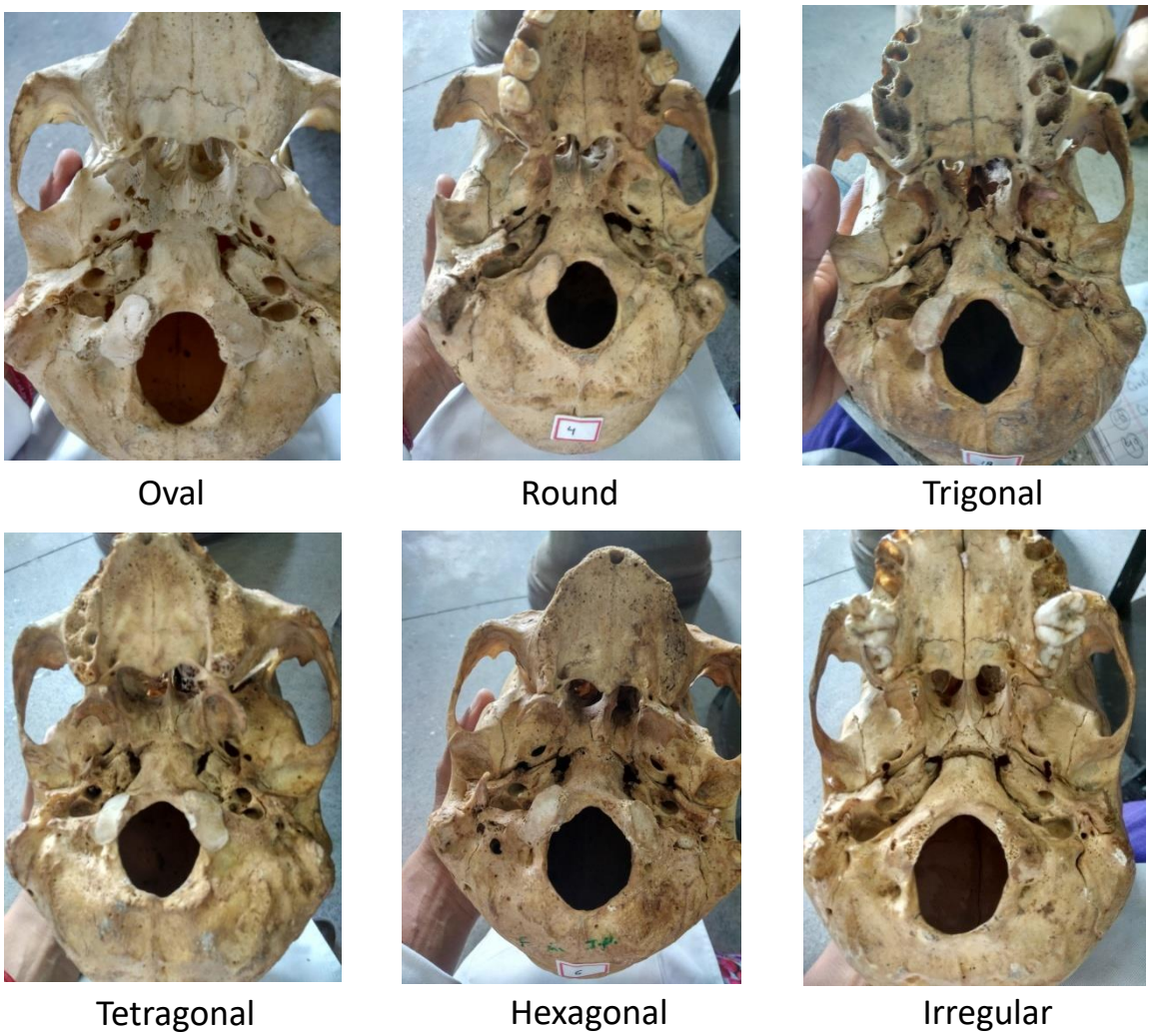


Fig. 2. Different shapes of foramen magnum

DISCUSSION

The knowledge of various morphometric parameters of the foramen magnum helps determine congenital malformations such as Achondroplasia and the Arnold-Chiari malformation, in which the shape and size of the foramen magnum vary in humans [4]. Patients with achondroplasia typically have an extremely small foramen magnum, whereas in cases of Arnold-Chiari malformation, there is an unusually large foramen magnum [4].

The "oval-shaped" foramen magnum was the most common shape in the current study. This shape was also the dominant shape observed in studies conducted by Kulesh et al. (2017) [5], Bharati et al. (2021) [6], Bharath et al. (2022) [7], and Gupta AK et al. (2022) [8]. However, Faazila et al. (2015) [9] found the egg-shaped (36%) foramen magnum to be the most dominant shape in their study. Rohini devi et al. (2016) [10] and Sarthak et al. (2017) [11] found the round shape to be the most common shape.

S. No.	Shapes of FM	AP diameter (mm) Mean ± SD	Transverse diameter (mm) Mean ± SD	FMI (mm) Mean± SD	FMA (mm^2) Mean± SD
2	Oval	35.0±3.1	28.7±2.2	78.8±28.0	826.7±84.0
2	Round	33.5±2.9	30.9±3.1	81.2±28.4	951.6±39.0
3	Trigonal	34.6±1.6	27.6±1.9	74.9±27.3	764.7±25.0
4	Tetragonal	32.4±2.5	29.0±3.2	73.8±27.1	868.2±77.0
5	Hexagonal	35.0±0.0	24.0±0.0	65.9±25.6	685.7±0.0
6	Irregular	34.6±1.8	26.0±1.0	70.6±26.5	778.7±43.0

Table 2. Dimensions of foramen magnum according to the different shapes

The second most common shape of the foramen magnum found was the "round shape," observed in 22.8% of skulls. These findings were similar to those of Bharati et al. (2021) [6], Gupta AK et al. (2022) [8], and Kulesh et al. (2017) [5]. The results of the current study were different from the findings of Faazila Fathima et al. (201) [9] and Bharath et al. (2022) [7], where the most common shape of the foramen magnum was "egg shape," while in our study, it was the round shape.

In the current study, a new uncommon shape, the "trigonal" shaped foramen magnum, was also observed in 5 skulls. Similarly, the uncommon shape of the foramen magnum was also reported by Archana et al. (2019) [12] and Giridhar et al. (2020) [13]. Archana et al. (2019) [12] found the "pear-shaped" foramen magnum in 8 skulls, and Giridhar et al. (2020) [13] found the "leaf-shaped" foramen magnum in 6% of skulls.

Total number (N) – 70	Antero-posterior (AP) diameter (mm)	Transverse Diameter (mm)	Foramen magnum Index (FMI)	Foramen magnum Area (FMA)(mm^2)
Mean ± SD	34.3±2.90	28.9±2.80	77.80±27.80	845.90±87.50
Minimum value	32.44	24.00	73.98	611.72
Maximum value	35.09	30.90	88.05	851.93

Table 3. Dimensions of foramen magnum of total sample (n = 70)

Sr. No.	Shapes of FM	Faazila Fathima *et. al*, 2015[9]	Kulesh S Chandekar *et. al*, 2017[5]	Archana *et.al*, (2019) [12]	Giridhar *et.al* (2020)[13]	Bharati *et al* 2021[6]	Gupta AK *et. al*, 2022[8]	Bharath et al. 2022[7]	Present study (2023)
1	Oval	26.42%	38.75%	33.3%	30%	35%	46.9%	36%	47.1%
2	Round	13%	32.5%	13.3%	12%	32.5%	18.8%	18%	22.8%
4	Trigonal	0	0	0	0	0	0	0	7.1%
5	Tetragonal	0	0	16.6%	0	25%	15.6%	8%	14.2%
6	Hexagonal	20.75%	0	16.6%	3%	7.5%	12.5%	6%	1.7%
7	Irregular	0	28.75%	0	27%	0	6.3%	0	7.1%
10	Egg	35.85%	0	0	17%	0	0	24%	0
11	Pentagonal	3.77%	0	13.3%	5%	0	0	8%	0
12	Leaf shaped	0	0	0	6%	0	0	0	0
13	Pear shaped	0	0	6.6%	0	0	0	0	0
Total		53	80			40	32	50	70

Table 2. Comparison of shape of foramen magnum with the result of other studies

In the current study, the mean anteroposterior diameter was 34.3±2.90 mm, and the mean transverse diameter was 28.9±2.80 mm. These findings were consistent with the findings of Archana et al. (2019) [12], Giridhar et al. (2020) [13], Bharat. J. Sarvaiya et al. (2018) [14], and M. Rohinidevi et al. (2016) [10] (refer to Table-5). Gruber P et al. (2009) [2] and Shikha Sharma et al. (2015) [3] measured higher anteroposterior and transverse diameters compared to our study (Table 5).

In the current study, the foramen magnum index was 77.80±27.80, which is lower compared to the findings of Bharat. J. Sarvaiya et al. (2018) [11] and M. Rohini Devi et al. (2016) [12]. The present study recorded significantly higher values of FMI (77.80±27.80) compared to Giridhar et al. (2020) [13] (1.21±0.12).

The mean foramen magnum area calculated in the present study was 845.90±87.50 mm^2. This value was similar to the values obtained by M. Rohinidevi et al. (2016) [10], whereas it was higher in comparison to the values by Giridhar et al. (2020) [13], Bharati et al. (2021) [6], and Bharat. J. Sarvaiya et al. (2018) [14]. Our recorded mean FMA was lower compared to Shikha Sharma et al. (2015) [3].

Limitations: The sample size of the present study was small and study done on dry human skulls of unknown age and gender.

CONCLUSION

Bony abnormalities of the craniovertebral junction are of interest not only to anatomists and physical anthropologists but also to surgeons, as they produce clinical symptoms that drastically affect human health.

S. No.	Authors	No.	Mean Anatero-Posterior diameter (mm)	Mean transverse diameter (mm)	Foramen magnum index	Foramen magnum area (mm^2)
1	Gruber P et al 2009[2]	110 skulls	36.6±2.8	31.1±2.7	-	-
2	Shikha Sharma et al 2015[3]	50 skulls	38.76	33.44	87.68	970.57
3	M. Rohinidevi et al 2016[10]	35 skulls	34.80	28.5	82.54	820.53
4	Bharat.J.Sarvaiya et al 2018[14]	326 skulls	34.18±2.74	28.49±2.13	83.60±6.21	766.86±104.76
5	Archana et al 2019[12]	120 skulls	33.79±2.60	28.25±1.83	83.91±6.43	-
6	Giridhar et al 2020[13]	64 skulls	34.10±2.63	28.07±1.87	1.21±0.12	752.07±111.97
7	Bharati et al 2021[6]	40 skulls	Male- 30±2.35 Female- 29.43±2.69	Male- 26.1±2.13 Female- 25.03±1.84	Male- 87.33±8.20 Female- 85.54±7.88	Male- 616.39±82.20 Female- 580.48±80.23
8	Present study	70 skulls	34.3±2.90	28.9±2.80	77.80±27.80	845.90±87.50

Table 3. Comparison of dimensions of foramen magnum with different studies

Abnormalities of the foramen magnum can be classified as congenital, developmental, acquired, traumatic, and pathological. These abnormalities can occur either alone or in combination. The data analysis and values of the current study may help anatomists, radiologists, and neurosurgeons with transcondylar surgical approaches, which are increasingly utilized for brainstem lesions and surgeries at the craniovertebral junction. Thorough knowledge of the anatomical variation of the foramen magnum helps radiologists differentiate deformities such as Arnold-Chiari malformation, in which the transverse diameter of the foramen magnum is increased.

These findings can also be useful to neurosurgeons for better approaches to treating foramen magnum meningiomas and other posterior cranial fossa lesions. The morphology and morphometry of the foramen magnum also have evolutionary importance. Further studies are needed, as there could be variations in the shapes and dimensions of the foramen magnum in different regions of India.

REFERENCES

1. Standring S. Gray's Anatomy. The anatomical basis of clinical practice. 41st ed. London: Elsevier Churchill Livingstone; 2016. p. 422.
2. Gruber P, Henneberg M, Boni T, Ruhli FJ. Variability of human foramen magnum size. Anat Rec (Hoboken). 2009;292(11):1713-19.
3. Sharma S, Sharma AK, Modi BS, Arshad M. Morphometric evaluation of the foramen magnum and variation in its shape and size: a study on human dried skull. Int J Anat Res. 2015;3(3):1399-403.

4. Richards G, Jabbour R. Foramen Magnum Ontogeny in Homo sapiens: A Functional Matrix Perspective. Anat Rec (Hoboken). 2011;294:199-216.
5. Kulesh S, Chandekar S, Jiwane NN. Morphometric study of foramen magnum of human skull in Vidarbha region. Int J Res Rev. 2017;4(5):157-60.
6. Bharati AS, Karadkhelkar VP, Zainuddin SS. Morphometric Study of Foramen magnum in East Godavari region of Andhra Pradesh. Int J Health Clin Res. 2021;4(5):294-97.
7. Bharath CNV, Priyadharsini S, Navajyothi D, Jyothi P, Guhan VN, et al. A study of morphometry and sexual dimorphism in foramen magnum in adult human skulls. Int J Sci Res. 2022;11(8):1-2. DOI: 10.36106/ijsr.
8. Gupta AK, Shah GJ, Chaudhary A, Prasad RJ. Foramen Magnum: A Morphometric Study in Dried Human Skulls. Nepal J Health Sci. 2022;2(1):1-6.
9. Faazila F, Yuvaraj Babu K. Evaluating the shape of foramen magnum and overlapping of occipital condyle on the foramen. Int J Sci Res. 2016;5(9):1078-82.
10. Rohinidevi M, Vimala V. Morphometric analysis of foramen magnum and variations in its shape in dried human adult skulls. Int J Anat Res. 2016;4(3):2670-73.
11. Sarthak J, Batham IK. Evaluation of foramen magnum in gender determination using helical CT scanning in Gwalior population. Int J Med Res Rev. 2016;4(3):357-60.
12. Singh A, Agarwal P, Singh A. Morphological and morphometric study of foramen magnum in dry human skull and its clinical significance. Int J Anat Radiol Surg. 2019 Jul;8(3):AO10-12.
13. Dasegowda G, Padmalatha K, Priyanka BP, Mirmire S. Morphology and morphometric analysis of foramen magnum in adult human skulls. Int J Anat Res. 2020;8(4.1):7771-76.
14. Sarvaiya BJ, Hathila SB, Chaudhari JS, Fichadiya NC. Morphometric analysis of foramen magnum in adult human dry skull of Gujarat region. Int J Anat Res. 2018;6(4.1):5787-02.

https://doi.org/10.46351/jas.v31i2pp141-149

JAS Journal of Anatomical Sciences (U.P. Chapter of Anatomical Society of India)

Original Article

SCULPTING LIFE: STUDYING THE INTRICACIES OF HUMAN FETAL SPLEEN MORPHOLOGY DURING DEVELOPMENT

A Shah[1], A K Singh[2], Richa Niranjan[3]

1. HNB Uttarakhand Medical Education University, Dehradun, India
2. Department of Anatomy, SSJ Government Medical College, Almora, India
3. Department of Anatomy, VCSG Government Medical College, Srinagar, India

ABSTRACT

Introduction: The spleen, a crucial lymphoid organ, plays a significant role in blood cell production and regulation. Emerging from mesodermal cells in the dorsal mesogastrium during the fourth embryonic week, the spleen is located in the left upper quadrant alongside the tenth rib. This study investigates morphological variations in human fetal spleens, serving as blood filters and storage sites for iron, erythrocytes, and platelets. This study aimed to assess morphological measurements of human fetal spleen development.

Materials and Methods: Sixty formalin-preserved fetuses (33 male, 27 female) of varying gestational ages were examined over a three-year period (2020-2023) at the Anatomy Department, Govt. Medical College Srinagar Garhwal, ensuring adherence to ethical guidelines. Gross characteristics such as position, shape, relations, notches & fissures, and ligaments were assessed and statistically analyzed.

Results: All fetal spleens were located in the left hypochondrium, with 50% exhibiting a wedge shape. The stomach consistently associated with the spleen, while no relationship was observed between the kidney, left colic flexure, and spleen in early fetal stages. The liver exhibited connections with the fetal spleen. Notches were most prevalent along the superior border, followed by the inferior border and lateral pole, with 27 spleens showing fissures.

Conclusions: Studying prenatal spleen development aids in understanding organ pathologies, thereby enhancing diagnostic and preventive techniques.

Keywords : Fetal Spleen, Wedge Shape, Notches, Fissures

Address for Correspondence:
Dr. Richa Niranjan, Professor & Head, Department of Anatomy, VCSG Government Medical College, Srinagar, India
Email: niranjanricha@yahoo.co.in

Date of Receiving: 27 Nov 2023
Date of Acceptance: 29 Dec 2023
ISSN: 0970-1842

INTRODUCTION

We delve into the mysteries of the spleen, a magnificent organ known as the human body's largest secondary lymphoid organ. This organ develops in the upper left quadrant of the abdominal cavity, between the fundus of the stomach and the diaphragm, and takes shape around the 5th week of intrauterine life as a mesenchymal condensation inside the dorsal mesogastrium [1,2].

At birth, the spleen weighs around 13g and has distinct surfaces and boundaries that contribute to its individuality. Its uneven visceral surface is defined by impressions from gastric, renal, pancreatic, and colic organs [2].

Surprisingly, studies on adult spleens show a 25% to 50% difference in spleen diameters between living and deceased individuals. This disparity is caused by variables such as blood drainage, decreased portal pressure, muscular contraction, and autolytic alterations [1].

The present work embraces this fascinating differential. This understanding leads us into a compelling investigation of the morphological development of the fetal spleen, revealing mysteries that bridge the gap between life and afterlife in the field of spleen related morphometrical and morphological research based on the various stages of gestational age.

MATERIAL AND METHODS

This study evaluated 60 fetuses (33 male, 27 female) with gestational ages ranging from 13 to 40 weeks; the study took place between the years 2020 and 2023. The specimens were obtained at the obstetric and gynecology department of the base teaching hospital Srinagar Garhwal, Uttarakhand. Fixed medically terminated fetuses were kept in 10% formalin. Prior to beginning the study, written agreements had to be obtained from the families, as well as approval from the institutional Ethical Board. Fetuses with obvious abnormalities or diseases were excluded from the research.

Sample Stratification: From the 13th to the 40th week of gestation. These 60 fetuses (33 male, 27 female) were divided into 3 groups: - Group I - 13-20 weeks (n=22), Group II - 21-30 weeks (n=18), and Group III - 31-40 weeks (n=20).

Gestational ages were calculated using a variety of criteria, including crown-rump length (CRL), biparietal diameter, head circumference, and foot length. Fig-1(A,B) Based on gestational age, the fetuses were divided into 3 groups: group 1 (13-20 gw), group 2 (21-30 gw), and group 3 (31-40 gw), respectively. Dimensional measurements were taken using an electronic weighing scale, digital vernier caliper, measuring tape, a plastic ruler, and saker tape. (gw-gestational weeks)

(A) **(B)**

Fig. 1. (A) Weight (wt) measurement of 28th GW fetus
(B) Head circumference (HC) of 32 GW fetus

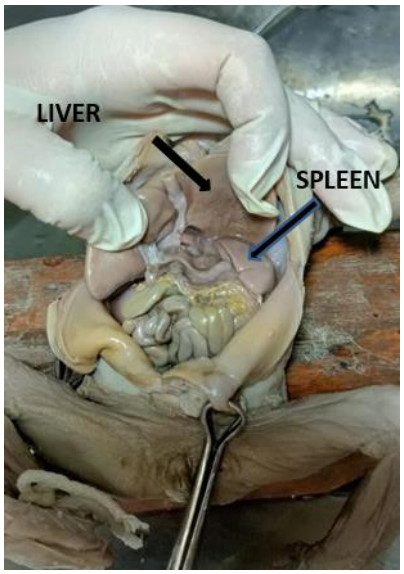


Fig. 2. Dissection of 32 GW fetus

(A) **(B)**

Fig. 3. (A) Length measurement of 32 GW fetal spleen
(B) Splenic notch on the superior border

Intra-abdominal tissues and the spleen were seen during abdominal dissection. The spleen's position in relation to the long axis, as indicated by the subcostal, vertical, and coronal planes, was studied. The relationships between the liver, stomach, colon, pancreas, kidney, adrenal gland, and diaphragm were investigated. The visibility of kidney and adrenal gland outlines was aided by the openness of the peritoneum at the posterior abdominal wall; otherwise, these organs were investigated by dissection. (Fig. 2)

The spleen's three primary ligaments—gastrosplenic, splenorenal, and phrenicocolic—were studied. The weights of the spleens were measured using an electronic weighing scale. Each spleen underwent a preparation step before weight and volume measurements were taken. (Table 2, 3)

The assessment of 60 dead human fetuses showed consistent features in spleen location and relationships with surrounding organs. Except at the hilum, [8,11] in all cases, stomach and spleen contact was made, with the spleen on the left and posterior to the stomach. In 95.5% of instances, the left colic flexure made contact with the spleen, mostly during the first trimester. Contact with the left kidney increased with gestational age. The spleen was continuously in touch with the left suprarenal gland, diaphragm, and tail of the pancreas.

The fetal spleens analyzed had a variety of forms, with the majority (65%) being wedge-shaped. In varying proportions, the stomach, left colic flexure, and pancreas were consistently connected to the fetal spleen. [Table-1] The study also looked at spleen notches and fissures, discovering the most amounts of notches on the superior border and fissures in 26 fetal spleens [3,4].

All parameters were defined using descriptive statistics. Using a one-way ANOVA test between and within groups, each morphological and metric was connected with separated gestational age groups. To detect sexual dimorphism, an independent sample test (T-test) was performed. This detailed observation gives vital insights into the fetal spleen's constant structural characteristics and alterations across gestational ages.

RESULTS

Shape of the fetal spleen	Numbers (n=60)	Percentage (100%)
Wedge shaped	30	**50%**
Irregular shaped	10	**16.666%**
Tetrahedral shaped	12	**20%**
Triangular shaped	5	**8.333%**
Oval shaped	3	**5%**

Table 1. Morphological features of fetal spleen

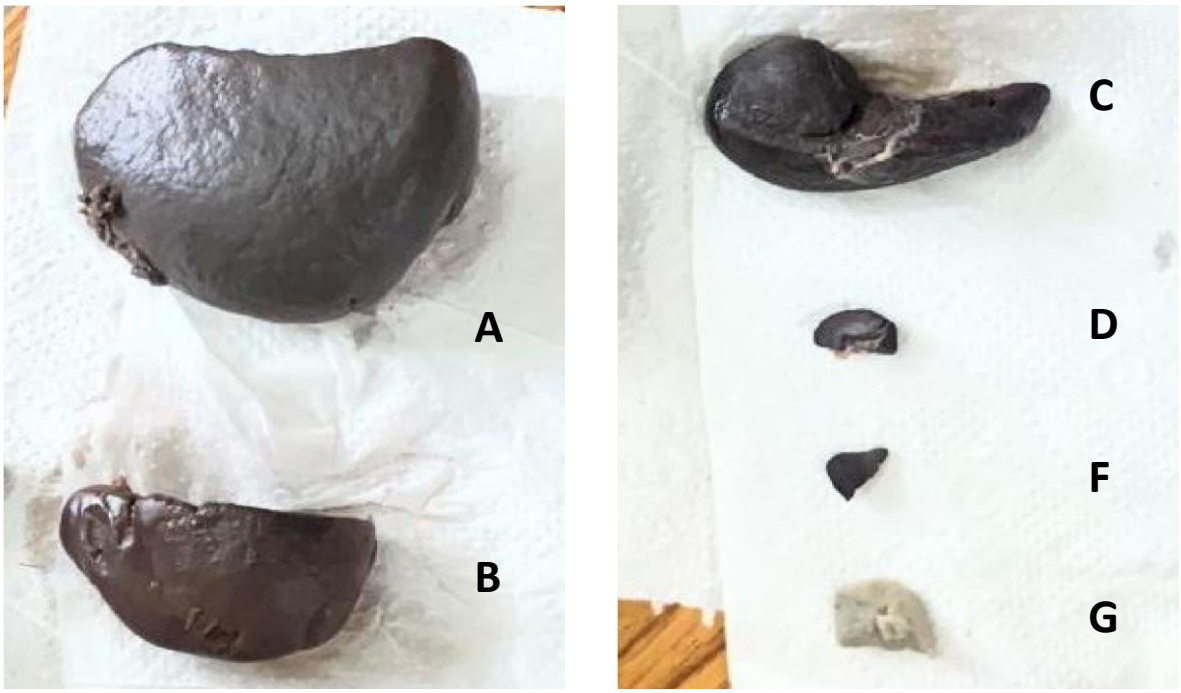


Fig. 4. Shapes of fetal spleen (A) Tetrahedral; (B) Wedge shaped; (C) Irregular shaped; (D) Oval shaped; (E) Triangular shaped; (G) Wedge shaped

GESTATIONAL WEEKS OF HUMAN FETUSES	MEAN±SD OF FETUS WEIGHT	MEAN±SD OF FETUS CHL	MEAN±SD OF FETUS CRL	MEAN±SD OF FETUS FL
GROUP1(13-20)	153.10±117.46	18.52±6.85	13.55±4.41	2.22±0.62
GROUP2(21-30)	854.34±339.79	36.58±4.22	25±4.07	5.49±1.05
GROUP3(31-40)	1940.32±339.72	46.77±1.64	31.14±0.88	6.90±0.70

Table 2. Mean weight, length, (CHL, CRL AND FL) of the human fetuses in different gestational age groups *(CHL-Crown Heel Length, CRL-Crown Rump Length, FL-Foot Length)

DISCUSSION

The present research focuses on the morphological development of the spleen throughout gestational weeks, with efforts to collect a larger sample from hospital resources. We were able to obtain 60 human fetuses, including gestational weeks 13 to 40, within the current limitations.

The average length, width, thickness, and weight of the fetal spleen across different gestational age groups demonstrated a steady rising trend with increasing gestational age. Further analysis revealed statistically significant changes in the mean fetal spleen length, width, and thickness across all gestational age ranges ($p < 0.001$).

GASTATIONAL AGE GROUPS	GROUP 1 (13-20WEEKS) (n=22)	GROUP 2 (21-30WEEKS) (n=18)	GROUP 3 (31-40WEEKS) (n=20)
MEAN WEIGHT OF FETAL SPLEEN(g) (mean ±SD)	1.78 ± 7.41	2.42 ± 2.03	4.66 ± 2.99
MEAN LEANGTH OF FETAL SPLEEN(cm) (mean ±SD)	1.05 ± 0.47	2.23 ± 0.79	2.93 ± 0.86
MEAN BREADTH OF FETAL SPLEEN(cm) (mean ±SD)	0.63 ± 0.28	1.51 ± 0.56	2.12 ± 0.51

Table 3. Mean weight, length, breadth, and thickness of the fetal spleen in different gestational age groups

Furthermore, a positive linear association was found between gestational age and spleen dimensions, emphasizing the complex interaction between fetal development and spleen morphology. This alignment with established observations from previous studies, such as Ungor et al. (2007), who carried out their study on 141 dead human fetuses aged between 9 and 40 weeks with no marked pathology and anomaly. The location of the spleen with neighboring structures, the existence of accessory spleens, notches on the border, fissures on the surfaces, major ligaments, and the shape of the spleen and its hilum were established.

The length, width, thickness, weight, volume, and the hilum dimensions of the spleen were measured. The dimensions, weight, and volume of the spleen increased with gestational age, and positive significant correlations were determined ($p < 0.001$). There was no difference between sexes in all parameters ($p > 0.05$). One or more accessory spleens have been found in 14% of cases.

Saheb et al. (2014) studied a total of 108 spleens collected from formalin-preserved fetuses. The measurements included length, width, thickness, and weight of the fetal spleen, and the ratio between fetal weight and spleen weight was measured after dividing the fetuses into 3 groups: 12-24 weeks, 25-36 weeks, and > 36 weeks. The average length, width, and thickness of the fetal spleen increased with gestational age, and the indices of splenic weight ranged between 0.33-0.35 percent. This reinforces the stability of the spleen location in the abdominal region.

Notably, during the embryonic stages of 6-10 weeks, the spleen was identified at the lumber level, later adopting its normal left hypochondriac position after 12 weeks, aligning with existing literature.

Examining morphometric aspects, measurements of length, width, thickness, and weight were conducted. Notably, the size of the spleen gradually increased from 1.5 cm at 13 weeks to 3.5 cm at 37 weeks gestation. The weight of the spleen exhibited a similar upward trend, ranging from 1.3 grams at 12 weeks to 9 grams at 38 weeks. These findings provide valuable insights into the developmental progression of the fetal spleen.

Regarding notches on the spleen borders, the superior border showed the highest incidence ratio (93.5%), followed by the inferior border (18%) and the lateral end or anterior extremity (7%). Notches were more numerous and deeper on the superior border, consistent with previous studies. The weight increase of the spleen was correlated with gestational age, aligning with the observations made by Henry Gray. Fissures were present in 32% of cases, primarily affecting the diaphragmatic surface, suggesting potential developmental defects or mechanical pressure influences. Interestingly, the relationship between the spleen and neighboring organs demonstrated variations from adult cases. The gastric impression and its consistent concave nature aligned with adults, while distinct rib impressions on the diaphragmatic surface were absent.

Adult spleen along with its anatomical variations has been studied extensively by several authors in human cadavers such as Das et al. (2008), Nayak et al. (2011), Rayhan et al. (2011), Chaware et al. (2012), Setty et al. (2013), Hussein et al. (2013), Chaudhari et al. (2014), Usha Kumari et al. (2014), Patil et al. (2014), Alex et al. (2015), Siva Chidambaram and Sridhar (2015), and Shankarrao and Rajendra (2016). They observed and counted notches on the borders of the spleen. The notches were mostly found on the superior border of the spleen in all the studies. Fissures on the diaphragmatic surface were also observed in some spleens. They also observed various shapes of the spleen such as wedge, tetrahedral and triangular, oval, scaphoid, V-shaped, liver-shaped. In the above studies, the wedge shape of the spleen was the most common.

In assessing relationships with surrounding structures, the left colic flexure, left adrenal gland, and pancreatic tail exhibited dynamic interactions during different trimesters. Notably, left kidney contact increased with gestational age. The study also delved into the presence of ligaments, with gastrosplenic and lienorenal ligaments consistently present and phrenicocolic ligament absent in a significant percentage, especially in the early fetal period. These detailed findings contribute to a comprehensive understanding of the morphological and relational aspects of the fetal spleen throughout various stages of development.

CONCLUSION

Studying the development of the spleen is essential given its dual role as a hematological and lymphopoietic organ during fetal life. Identification and palpation of notches on the superior border play an important role in diagnosing splenomegaly, providing a helpful foundation for clinicians in clinical practice. Furthermore, investigating the interactions between the spleen and other organs adds to our understanding of their development throughout fetal life. The lack of literature on the morphological evolution of the spleen emphasizes the significance of this research. It not only lays the groundwork for additional study, but it also has potential uses in pediatric medicine and surgery. This work adds to our understanding of spleen morphology, providing useful insights for both medical research and practical applications.

REFERENCES

1. Gray A. Gray's Anatomy. Spleen: The Anatomical Basis of Clinical Practice. 40th ed. Elsevier Churchill Livingstone; 2008. Edinburgh. pp. 1239-1244.
2. Moore KL, Persaud TVN, Torchia MG. The Digestive System. The Developing Human: Clinically Oriented Embryology. 8th ed. Elsevier publication; 2008. New York, New Delhi. p. 224.
3. Langman's Medical Embryology. 12th ed. Wolters Kluwer, Lippincott Williams & Wilkins; 2012. Philadelphia. p. 213.
4. Schoenwolf BC, Bleyl SB, Brauer PB, Francis West PH. Spleen in: the Development of the Gastrointestinal Tract.
5. Larsen's Human Embryology. 9th ed. Elsevier Churchill Livingstone; 2009. Philadelphia. p. 454.
6. Singh R, Kumari G. Morphological assessment of human fetal spleen. Asian J. Biomed. Pharmaceut. Sci. 2016;6(52):19-21.
7. Ferreira GF, Rega GM, Mandarim de Lacerda CA. Spleen's relative growth in human fetuses. Bull. Assoc. Anat. 1992;76(233):23-25.
8. The abdominal cavity. Cunningham Manual of Practical Anatomy. 15th ed. Oxford University Press; 2008. New York. pp. 126-127.
9. Üngör B, Malas MA, Sulak O, Albay S. Development of spleen during the fetal period. Surg. Radiol. Anat. 2007;29:543-550.
10. Sadler TW. Langman’s Medical Embryology. 12th ed. Wolters Kluwer, Lippincott Williams & Wilkins; 2012. Philadelphia. pp. 213.
11. Ajmani. Practical embalming. Textbook of ‘Embalming principles and legal aspects’. 1st ed. New Delhi: Jaypee publishers; 1996. pp. 131-135.
12. Saheb H, Velichety SD, Haseena S. Morphological and Morphometric study of Human Foetal Spleen. Int J Anat Res. 2014;2(1):234-238.

13. Mukhia R, Mukherjee A, Sabnis A. Histogenesis of Human Fetal Spleen. Int J Anat Res. 2016;4(1):2119-2124.
14. Moore KL, Dalley AF, Agur AMR. Spleen in Abdomen. Clinically oriented anatomy. 6th ed. New Delhi: Wolters Kluwers (India) Pvt Ltd; 2010. pp. 263-265.
15. Pal M, Naranbabu Singh TH, Rajendra Singh CH. Histogenesis of spleen in human fetuses. Journal of the Anatomical Society of India. 2013;62:139-145.
16. Mebius RE. Organogenesis of lymphoid tissues. Nature Reviews Immunology. 2003;3(4):292-303.
17. Bain BJ. Spleen and red blood cells. Blood Cells: A Practical Guide. 4th ed. Wiley-Blackwell; 2006. pp. 171-188.
18. Gruver AL, Sempowski GD. Cytokines, leptin, and stress-induced thymic atrophy. Journal of Leukocyte Biology. 2008;84(4):915-923.
19. Sharma S, Saxena R. Human fetal spleen and spleen colony-forming units. Clinical and Developmental Immunology. 2005;12(2):117-120.
20. Das P, Chakraborty D. Development of the human spleen during prenatal life. The Journal of Obstetrics and Gynecology of India. 2016;66(1):39-45.
21. Lakshminarayana KG. Immunohistochemical study of the human fetal spleen. Journal of Clinical and Diagnostic Research. 2008;2(6):1066-1071.
22. Sarkar S, Kar SK. Functional significance of the fetal spleen. Medical Journal Armed Forces India. 2015;71(Suppl 1):S36-S39.
23. Rajakumar S, Vadivel S. Development of human spleen. Indian Journal of Pathology and Microbiology. 2003;46(4):596-598.

https://doi.org/10.46351/jas.v31i2pp150-154

JAS Journal of Anatomical Sciences (U.P. Chapter of Anatomical Society of India)

Original Article

UNVEILING THE IMPACT: ANATOMY INTERNAL ASSESSMENT EXCLUSION IN MBBS FIRST PROFESSIONAL RESULTS

Sushobhana[1], Shweta Singh[1], Ranjana Sachan[2], Abeer Zubair Khan[1], Dewanshi Mishra[3], Pratibha Dwivedi[1], Kirti Gupta[1]

1. Department of Anatomy, Hind Institute of Medical Sciences, Safedabad, India
2. Department of Anatomy, Autonomous State Medical College and Hospital, Hardoi, India
3. Department of Anatomy, Dr. Ram Manohar Lohia Institute of Medical Sciences, Lucknow, India

ABSTRACT

Introduction: In 2019, the National Medical Commission (NMC) implemented the Competency-Based Medical Education (CBME) curriculum, altering teaching, learning, and assessment methods. Internal assessment marks, once contributing to final grades, now only serve as eligibility criteria. This study, conducted at the Department of Anatomy, HIMS, Safedabad, examines the impact of excluding internal assessment from MBBS 1st year students' final Anatomy marks.

Materials and Methods: This study assessed the performance of MBBS 2019 (CBME) and MBBS 2018 (old) batches in Anatomy. Parameters examined included: 1) first attempt exam clearance rates, 2) passage via grace marks, and 3) distribution of scores (students who scored >70%, between 60-70% and between 50-60%). Statistical analysis employed chi-square tests.

Results: In the MBBS 2018-19 batch, 86 out of 100 students passed Anatomy, compared to 94 out of 99 in the MBBS 2019 (CBME) batch. In the MBBS 2018 batch, excluding internal assessment would have resulted in 44 failures, reduced to 14 with inclusion. Chi-square tests demonstrated significant differences in scores and failure rates.

Conclusions: Excluding internal assessment markedly affected Anatomy exam outcomes, highlighting its pivotal role in MBBS 1stProfessional success.

Keywords : Competency-Based Medical Education (CBME), Internal assessment, Anatomy, Medical education, MBBS curriculum, Student performance

Address for Correspondence:
Dr. Sushobhana, 1- Assistant Professor, Department of Anatomy, Hind Institute of Medical Sciences, Safedabad, India
Email: sushobhana@gmail.com

Date of Receiving: 30 Nov 2023
Date of Acceptance: 31 Dec 2023
ISSN: 0970-1842

INTRODUCTION

Assessment is derived from the Latin word “assidere,” meaning “to sit with.” Therefore, it is a collaborative process between educators and students, not something done to students [1]. Competency-based curriculum assesses learning longitudinally to identify needs, plan remedial measures, and provide learning opportunities [2]. An effective system of internal assessment is one that not only evaluates the knowledge of the learner but also the process by which it is acquired.

In August 2019, NMC introduced a competency-based undergraduate curriculum and adopted the longitudinal process to assess all competencies, thereby changing the teaching and learning method as well as the method of assessment [3,4]. Before 2019, the marks of internal assessment scored by students were previously added to the final award list. Now, from 2019, the internal assessment serves as an eligibility criterion only. It is not added to the final score but is reflected separately in the final mark sheet [4].

MATERIAL AND METHODS

This study was carried out in the Department of Anatomy at HIMS, Safedabad, Barabanki. The marks obtained by the students of the MBBS 2018 batch (old batch - it followed the traditional method of assessment) and the MBBS 2019 batch (CBME batch - it followed the new CBME curriculum batch) were taken.

It was conducted to determine the effect of not including internal assessment marks in the final marks of MBBS 1st-year students in the subject of Anatomy. There were 100 students from the MBBS 2018 (old) batch and 99 students from the MBBS 2019 (CBME) batch.

The effects of this change in the method of assessment were evaluated based on the following criteria:

1. Percentage of students who cleared the exam on the first attempt in the MBBS 2019 (CBME) batch vs. MBBS 2018 (old) batch.
2. Percentage of students who finally passed through grace marks.
3. Number of students who scored >70%, between 60-70%, and between 50-60%.
4. Percentage of failed students in the MBBS 2018 (old) batch who passed due to the inclusion of internal assessment marks in the final award list. The above parameters were tabulated in a Microsoft Excel sheet. Statistical analysis was performed using the chi-square test, and the p-value was determined.

RESULTS

More students passed in the MBBS 2019 (CBME) batch than in the MBBS 2018 (old) batch. In the first attempt, 95% of students passed in the MBBS 2019 (CBME) batch. Nine students scored more than 70% marks,

46 students scored between 60% to 70%, and 39 students scored between 50% to 60% marks. (Table 1)

In the 2018 (old) batch, only 86% of students passed in the first attempt. Only one student scored >70% marks, 20 students scored between 60% to 70%, and 65 students scored between 50% to 60% marks. (Table 1)

After re-evaluation, two students passed by grace marks in the MBBS 2019 (CBME) batch, taking the total pass percentage to 97%. In the MBBS 2018 (old) batch, seven students passed by grace marks, but seven students still failed. The total pass percentage now was 93%. (Table 1)

In the 2018 (old) batch, 45 students would have failed if the internal assessment had not been included in the final score of the assessment. When the internal assessment was included in the final score, only seven students failed, and the rest seven passed by grace marks. (Table 2) Statistical analysis of the number of students scoring more than 60% and between 50% to 59% in both the 2018 (Old) and 2019 (CBME) batch showed the chi-square test value as 25.969 with two degrees of freedom. The p-value was 0.00001, which showed a highly significant difference between the scores of both batches and a statistically significant difference between the number of students who failed. (Table 3)

MBBS Batch	2018-19(old batch)	2019-20 (CBME)
No. of students appeared	100	99
No. of students passed in 1st attempt	86	94
Percentage of students passed	86%	95%
No. of student with score >70%	1	9
No. of students with score between 60-70%	20	46
No. of students with score between 50-60%	65	39
No. of students who passed due to grace mark	7	2
No. of students Failed	7	3
Total pass percentage	93	97

Table 1. Outcomes of assessment of MBBS 2018 (old) and MBBS 2019 (CBME) batch

MBBS 2018 (Old) batch	
Total no. of students	100
No. of students who passed	93
No. of students Failed (supplementary)	7
No. of students who passed due to grace marks	7
No. of student who would have failed if internal assessment marks was not included in the final award list	45
Percentage of failed students who were saved due to inclusion of internal assessment marks	84.44%

Table 2. Effect of inclusion of internal assessment marks on outcome of MBBS 2018 (old)

	MBBS 2018 (old) batch	MBBS 2019 (CBME) batch	Chi Square Test, df	p-Value
No. of students with score >60%	21	55	25.969, 2	0.00001
No. of students with score between 50% to 59%	65	39		
No. of students failed	14	5		

Table 3. Categories of students based on performance and their statistical analysis

DISCUSSION

The change in the method of assessment in the 2019 (CBME) batch is evident from the fact that the pass percentage was better in the 2019 (CBME) batch than the 2018 (old) batch, which did not follow the CBME curriculum. Out of 99 students of the 2019 (CBME) batch, 97 passed and were promoted to the second professional, and out of these, 55 students scored more than 60% in the first attempt in the first professional examination.

In this 2019 (CBME) batch, internal assessment was conducted throughout the year using different assessment methods for assessing different competencies. Internal assessment was conducted through various modes of online and offline examinations, and assignments were given after the completion of each topic. Both online and offline modes of teaching were used, along with demonstration videos shared for revision. Constructive feedback was provided, which improved the students' learning. Teachers were able to guide the students to take remedial measures on time before the final exam [5]. It empowered them to understand and learn in a stress-free environment [4]. Teachers were able to gather more information on students' learning levels and their progress through logbooks, manuals, and seminars [6].

Out of 100 students, 93 students passed and were promoted to the second professional, and only 21 students could score >60%. In the traditional method, when the internal assessment was added to the final score, students were more stressed and anxious to score in the internal assessment throughout the year. The focus was more on scoring, and there was less involvement of students in self-assessment and reflections [4]. The results of our study indicated not only a better pass percentage but also a better score in the MBBS 2019 (CBME) batch. This contrasts with a study done in the Pharmacology Department at GMC, Jalgaon, Maharashtra,

where the MBBS 2018 (old curriculum) batch performed better [7].

CONCLUSION

In the MBBS 2018 (old) batch, internal assessment was added to the final score, but fewer students were able to score >60%. Internal assessment in the MBBS 2019 (CBME) batch was important as a formative assessment. This helped the students to perform at an optimal level in the first professional examination. More students passed with better scores. This study pertains to the first batch of CBME. More studies on upcoming batches should be conducted to gain a better understanding of the benefits and disadvantages of internal assessment.

REFERENCES

1. A Short Glossary of Assessment Terms. [Internet]. [cited 2020 Feb 15]. Available from: https://serc.carleton.edu/introgeo/assessment/glossary.html.
2. Frank JR, Snell LS, Cate OT, Holmboe ES, Carraccio C, Swing SR, et al. Competency-based medical education: Theory to practice. Med Teach. 2010;32:638–45.
3. Badyal DK, Singh T. Internal assessment for medical graduates in India: Concept and application. CHRISMED J Health Res. 2018;5:253-8.
4. Medical Council of India. Assessment Module for Undergraduate Medical Education Training Program, 2019. 1-29.
5. Badyal DK, Singh S, Singh T. Construct validity and predictive utility of internal assessment in undergraduate medical education. Natl Med J India. 2017;30:151-54.
6. Fitzgerald JT, John B, Steven K, Patricia M, Santen AS, Kent JS, et al. Assessment challenges in competency-based education: A case study in health professions education. Med Teach. 2015;38:1-9.
7. Sharp K, Teli SE, Mahajan H, Sonawane R. Comparative study between academic performances of traditional versus competency-based medical education M.B.B.S. curriculum students in Pharmacology. Natl J Physiol Pharm Pharmacol. 2023;13(Online First). DOI: 10.5455/njppp.2023.13.09461202202102022.

https://doi.org/10.46351/jas.v31i2pp65-81

JAS Journal of Anatomical Sciences (U.P. Chapter of Anatomical Society of India)

Original Article

CHALLENGES IN REPRODUCING THE STANDARD LITHIUM PILOCARPINE MODEL OF TEMPORAL LOBE EPILEPSY IN *WISTAR* RATS: TROUBLESHOOTING STRATEGIES

Asha Rao[1], Tulika Gupta[1], Ashish Kumar Kakkar[2], Shammy Chandel[3], Ashish Jain[2], Lekha Saha[2], Ranjana Bharti[1], Ujjwaljit Kaur[1], Navneet Singla[3]

1. Department of Anatomy, PGIMER, Chandigarh, India
2. Department of Pharmacology, PGIMER, Chandigarh, India
3. Department of Neurosurgery, PGIMER, Chandigarh, India

ABSTRACT

Introduction: Animal model of temporal lobe epilepsy (TLE) provide an essential platform to study the disease pathophysiology, and disease modifying management protocols. Lithium –pilocarpine model is one of the most favored model. Many practical difficulties have been encountered while creating this model in this study as well as by other authors. We have systematically attempted to compile all issues and their possible solutions in establishment of Li-Pi model of status epilepticus in *wistar* rats, to provide a dependable protocol with improved seizure induction rates and lower mortality rates.

Materials and Methods: Young small size male wistar rats (100-150gm) were injected with lithium chloride (LiCl) (127 mg/kg, i.p.), followed by methyl scopolamine (1mg/kg, i.p.) after 18-22 hrs. Precisely after 30 minutes, dose of pilocarpine (i.p.) was administered. Post seizures, diazepam (10 mg/kg) was injected to reduce seizures severity and to increase survival rate and rats were fed glucose (10% of body weight) through oral gavage. Model creation was validated by histological examination of hippocampus. The rats which did not develop seizures, were reutilized for the model generation after drug wash-out period of 48 hours.

Results: Initial model creation was unsuccessfully attempted in 75 rats. Standard described protocol of three incremental doses of pilocarpine (10mg/kg, maximum of 30mg/kg) did not elicit epilepsy. Loading dose of 30mg/kg pilocarpine caused stage 1 epilepsy in 60% rats; second dose of pilocarpine (30mg/kg), caused stage 2 epilepsy in 70% rats. Successful TLE model creation was seen in 33 out of 57 rats, using proposed modification in which 2 incremental doses of pilocarpine (30mg/kg each) were followed by 3 doses of 10mg/kg each; 6 rats died.

Conclusions: The study provides a detailed procedure for lithium pilocarpine model creation and various pitfalls encountered. Modified pilocarpine dosage led to improved seizure induction rates (58%) and lower mortality rates (10%).

Address for Correspondence:
Dr. Tulika Gupta, Additional Professor, Department of Anatomy, PGIMER, Chandigarh, India - 160012
Email: tulikag11@gmail.com Phone: 0172 2755272

Date of Receiving: 09 Nov 2023
Date of Acceptance: 20 Dec 2023
ISSN: 0970-1842

Keywords: Epilepsy animal model; Status epilepticus; lithium-pilocarpine; Animal mortality

INTRODUCTION

The World Health Organization states that epilepsy is a chronic non-communicable disease characterized by recurrent seizures, sometimes accompanied by loss of consciousness and loss of control of bowel or bladder function. Status epilepticus (SE) is a persistent seizure lasting for more than five minutes, causing long-term detrimental consequences [1]. In India, the prevalence rate for epilepsy varies from 1.2 to 11.9 per 1000 population among adults [2, 3]. Epileptic patients experience a reduced quality of life, an increased risk of psychosocial dysfunction, injuries, and premature death [4-6]. Therefore, there is an imperative need to understand the pathophysiology of epilepsy in order to develop preventive or disease-modifying therapies. This need has led to an intense research focus on epilepsy. Epilepsy research is conducted either on human tissue samples obtained from surgery on epileptic patients or on animal models of epilepsy [7-10]. Human surgical tissue has two drawbacks: it cannot be used to study acute changes during epileptogenesis as surgery is performed on chronic and drug-resistant patients, and interventional studies are not possible.

Animal models of epilepsy have proven to be of immense importance. Animal models allow for detailed study of the disease pathophysiology. Interventions can be performed for experimentation as well as validation, thus allowing testing of the efficacy, mechanism of action, and side effects of potential therapeutic agents [11]. An ideal animal model of epilepsy should have disease mechanisms as well as phenotypic features similar to human epileptic conditions; moreover, it should demonstrate predictive validity, representing the treatment responses observed clinically [12, 13].

Chemoconvulsants and Electrical (Kindling) are the two most commonly used animal models to study mesial temporal lobe epilepsy (MTLE) [14, 15]. Chemoconvulsant models of epilepsy are created by administering excitotoxic substances and include the Kainic acid model [16-22] and the Lithium-pilocarpine (Li-Pi) model of epilepsy [14, 23]. We chose the Li-Pi model to study the role of calcium channels in epilepsy-induced cell death through an interventional study.

The Li-Pi model replicates the natural history of human MTLE. Similar to human temporal lobe epilepsy, the seizure focus is localized in the limbic areas [24], interictal activity is generated in the subiculum [25], upregulation of neurotrophins has been noted in the hippocampus [26], and pathologically, granule cell dispersion, cell death, and mossy fiber sprouting are observed [27]. Lastly, cognitive and memory impairment is noted [28, 29], with an initial epileptogenic insult in the form

of status epilepticus leading to the acute phase of MTLE. In the kainic acid epilepsy model, there are certain limitations; unlike the pilocarpine model, the stages of seizures are not easily distinguishable, and the animals show behavioral changes such as anxiety and depression.

Although the Li-Pi model has become a favorable model for conducting epilepsy-related studies, it has a high mortality rate [30-33]. We encountered many difficulties while replicating the standard Li-Pi model. Similar issues have also been reported by many studies [34, 35]. Thus, here we have attempted to provide detailed steps for the creation of the Li-Pi model of TLE in *Wistar* rats. Issues faced and their possible solutions have been addressed with the aim of creating a dependable protocol with improved seizure induction rates and lower mortality rates.

MATERIAL AND METHODS

Selection of animals

Seventy-five (n=75) male Wistar rats weighing 100-150g were obtained from the Central Animal House facility at the Post Graduate Institute of Medical Education and Research (PGIMER), Chandigarh. All experimental protocols were approved by the Institutional Animal Ethics Committee (IAEC no. 803/116th dated 14th September 2021). Clear plastic shoebox cages containing corn cob bedding were used to house two rats per cage, under a 12-hour light: 12-hour dark cycle to acclimatize rats to the light/dark cycle, at a constant temperature (22 ± 2 °C) with free access to food and water. Rats were acclimatized for at least one week before being used in the experiments. Extreme care was taken to minimize the pain and discomfort of the animals through interventions (Fig. 1).

Fig. 1. Acclimatization of rats (2rats/cage)

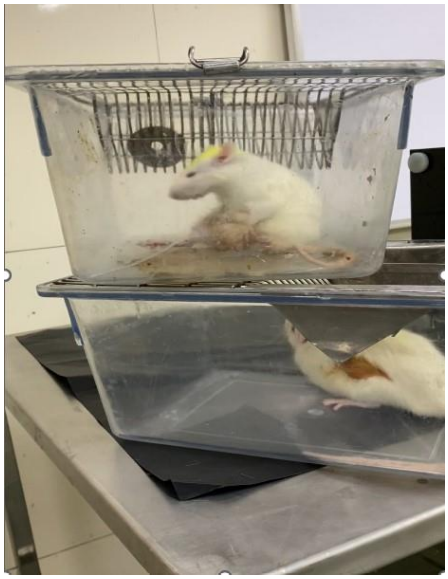


Fig. 2. Still image from video 1

LPISE rat model

For model establishment, pilocarpine hydrochloride, methyl scopolamine, lithium chloride (LiCl) were purchased from Sigma-Aldrich, and diazepam, ketamine, and xylazine were procured from a medical shop. Prior to the initiation of any experiment, the weight of the rats was recorded. Then, the rats were segregated into two groups, namely, control and epilepsy groups.

Standard Method of Model Development: As per the literature, LiCl (127mg/kg), methyl scopolamine (1mg/kg), and pilocarpine (30mg/kg) induce SE in rats via the intraperitoneal (i.p.) route [36-38]. In our study, we faithfully tried this protocol in 75 rats. LiCl was administered at a dose of 127mg/kg (18-22 hrs before pilocarpine administration), and the next day, 30 minutes before pilocarpine dosage, methyl scopolamine (1mg/kg) was given to decrease the peripheral effects of pilocarpine. During several attempts, pilocarpine was given at a dosage of 30mg/kg intraperitoneally, either as a single dose or as a breakup of a 30mg/kg dose (10+20mg/kg or 15+15mg/kg); however, SE was not observed even a single time.

Continuous video monitoring was done to observe the same. There was no mortality. Later, we came across a paper [34] in which the authors recommended the subcutaneous route for pilocarpine (25mg/kg to 75mg/kg) with low mortality (up to 1%) [34]. So, we tried to reproduce the same but achieved success only at a higher pilocarpine dose (90 mg/kg). Comparison revealed that seizures came earlier and were of greater intensity when pilocarpine was injected intraperitoneally compared to subcutaneously, at the same dose.

Modified Method of Model Development: After the failure of the method given in the literature, for model development, we used young small-sized rats (100-150g). In the epileptic group, LiCl (127 mg/kg) was injected via the intraperitoneal route. After 18-22 hrs, methyl scopolamine (1mg/kg), a muscarinic acetylcholine receptor antagonist, was injected intraperitoneally to decrease the peripheral effects of pilocarpine. Precisely after 30 minutes, two incremental doses of pilocarpine (30mg/kg each) were followed by two doses of 15mg/kg each until the occurrence of seizures (a maximum dose of 90 mg/kg of pilocarpine) via the intraperitoneal route.

The rats developed seizures at a high dose of pilocarpine (80-90 mg/kg). Once the rats started to develop seizures, diazepam (10mg/kg) was injected to reduce seizure severity. To increase the survival rate, glucose was orally fed (10% of the body weight) to rats through oral gavage, and their eyes were kept moist by swiping them with water-soaked cotton swabs to prevent dehydration in rats. In concordance with the

3Rs (Replacement, Reduction, and Refinement) of animal welfare use, rats that did not develop seizures even after the maximum dose of pilocarpine were reused for model development after a washout period of 48 hours; about half of these rats developed seizures.

Video monitoring and seizure analysis

During model development, a blinded observer rated the seizure severity using a modified Racine scale (Salem, El-Shamarka et al., 2018, [39]):

• Stage 1: Staring with mouth clonus
• Stage 2: Head nodding, automatisms (e.g., scratching, sniffing orientation)
• Stage 3: Unilateral forelimb clonus
• Stage 4: Forelimb clonus (rearing)
• Stage 5: Forelimb clonus with rearing and one fall
• Stage 6: Forelimb clonus with rearing and multiple successive falls
• Stage 7: Tonic/clonic seizures (e.g., running and jumping)

SE was assumed to occur in rats with seizures of stage 4 and above (Video 1: [link]) (Fig. 2). The time of SE arrest, and 24-hour survival rate were noted. The weight of animals was taken consecutively for 3 days; starting from the day when LiCl dosing was started, the next day before giving methyl scopolamine, and then on the third day when they were sacrificed after 24 hours of *model development. Their weight was found to be decreased by 7-10g on the third day.*

Tissue harvesting

After the generation of the epilepsy model, the rats that developed seizures were selected for downstream processing. Rats were anesthetized using ketamine (100 mg/kg i.p.) and xylazine (10 mg/kg i.p.) [40] and then sacrificed by cervical dislocation. The brain was harvested, and the hippocampus was isolated and stored in 10% buffered formalin, RNA later, and frozen at -80 degrees for histology, gene expression, and protein studies, respectively. Sacrificed rats were disposed of in yellow polybags for incineration.

Histological validation of Temporal lobe epilepsy

It was done by identifying two hallmark features – Granule cell dispersion (GCD) and cell death in the hippocampal tissue of rats in Nissl-stained slides [41]. In all rat epilepsy cases (n=18), statistically significant GCD and cell death were observed in comparison to the control rats. (Fig. 3).

RESULTS

Results with standard protocol:

The standard protocol was attempted eight times (8 animals – 6 epilepsy group,

* Video link at the end of the article

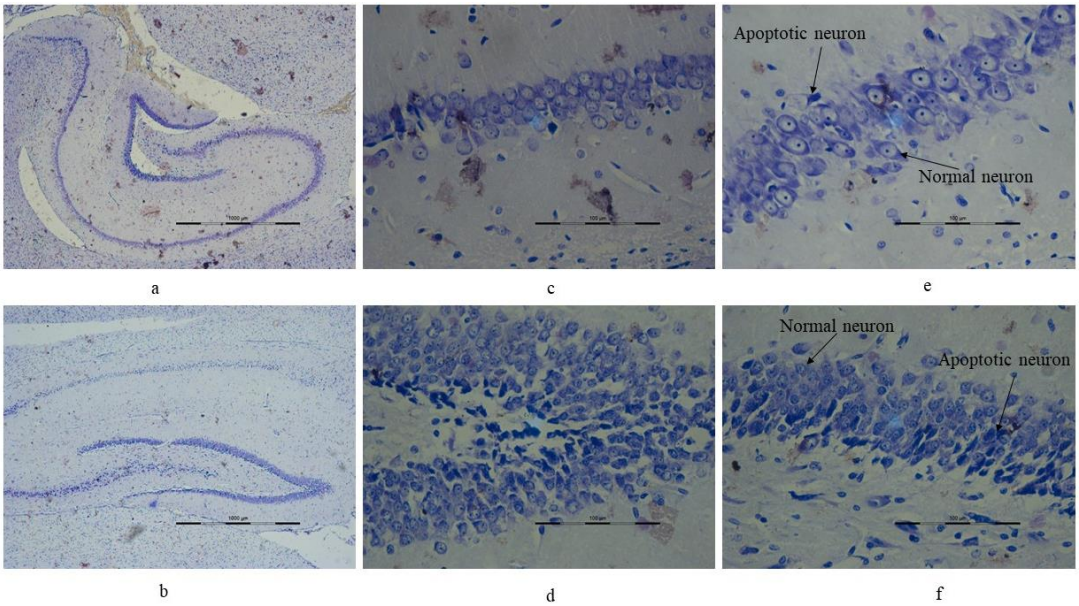


Fig. 3. Results of Nissl's staining showing- a. & b. Hippocampus at 4X in control rat (a.) and epileptic rat (b.) c. & d. GCD seen in epileptic rat (d.) as compared to the compact granule cell layer in control rat (c.), at 40X. e. & f. Extensive neuronal apoptosis seen in the epileptic rat (f.) as compared to control rat (e.) hippocampus at 40X.

2 control group) with all due precautions, but none of the rats demonstrated status epilepticus. There was no mortality. In one batch, LiCl was administered at a double dose (254mg/kg), but still, the rats did not experience seizures.

Results with modified protocol

For every batch, six rats were used for model development, and two rats were taken as controls. Most of the time, out of these 6 rats, 2 rats developed seizures of stage 4 and above, 2 rats were moderately sensitive to the increased pilocarpine dose, i.e., they showed Racine scoring stage of <4. 2 rats were completely resistant to the induction of epilepsy despite the increased pilocarpine dose (90mg/kg) (Fig. 4). A total of 75 rats were utilized for the model establishment. These rats were divided into 9 sub-groups; 18 rats were in the control group, and 57 rats were in the epilepsy group. Fate of 57 epileptic rats: 18 rats (31.57%) developed seizures of stage 4 and above; 15 rats (26.31%) were moderately sensitive (less than stage 4), remaining 18 (31.57%) rats were resistant to the increased dose of pilocarpine, and 6 rats died (10.52%). Deaths occurred within 24 hours of the model development. (Fig. 5)

Complications encountered

1.Resistance to Model Development: Some rats didn't develop seizures (31.57%). Even after the maximum dose of pilocarpine, no external change was visualized.

2. Shivering: In some batches of rats, even at a dose of 90 mg/kg, they didn't enter stage 4 and above of seizures, but they were shivering, though the same was not visible externally in terms of forelimb clonus, rearing, and falling. Shivering was not classified as any stage of seizures. Since the rats were weak and shivering, further doses

were not given to them.

3. Moderately Sensitive: On following every possible troubleshooting advice, we found that the maximum number of rats were only moderately sensitive to the dose of pilocarpine being given (90 mg/kg) and exhibited seizure severity less than stage 4.

4. Alopecia & Patchy Skin: In a single case, a rat suffered from the loss of body hair on the ventral side and yellowish patchy skin. (Fig. 6)

DISCUSSION

We attempted to generate the LPISE model of TLE to investigate the role of the mitochondrial calcium uniporter in an animal model of SE. Being a stable model of TLE, this model is useful for studying both acute and chronic epileptic changes. Moreover, studies on etiopathophysiology and management of epilepsy using various interventions can be performed on this model.

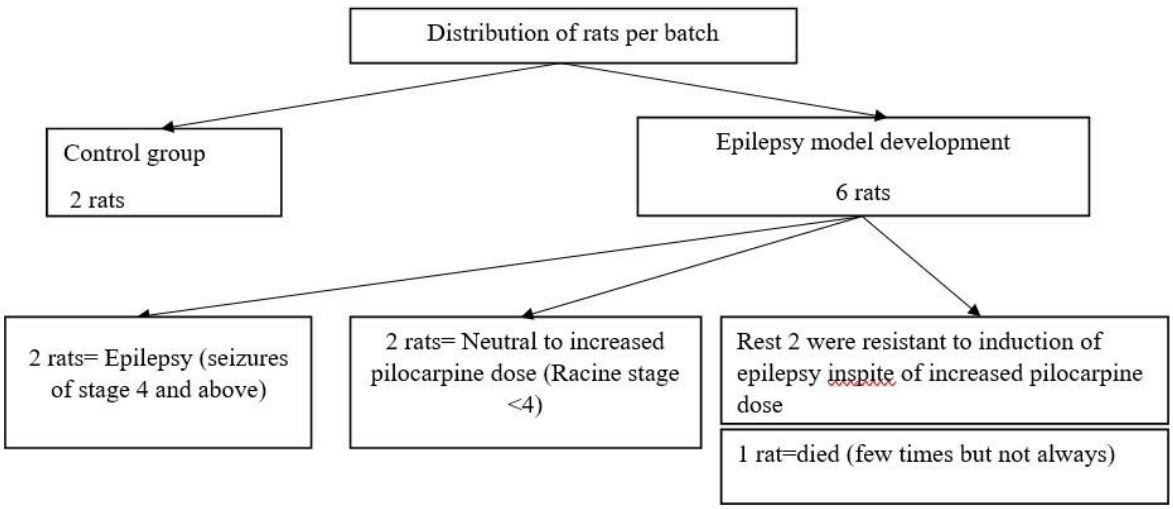


Fig. 4. Flow chart showing distribution of rats per experimental batch

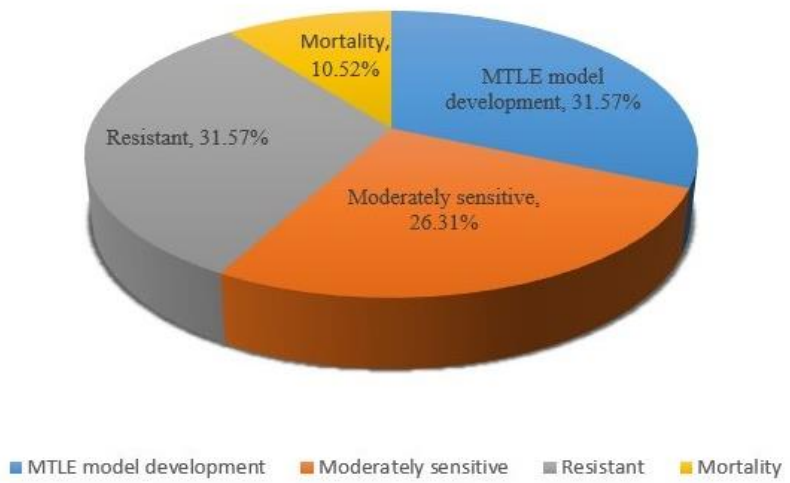


Fig. 5. Pie-chart showing response of rats towards lithium-pilocarpine

Fig. 6. Various complications encountered during model development. a: Alopecia; b: Patchy skin.

According to many reports, this epilepsy model can be created with ease [36, 37, 42]. Contrary to this, we could not achieve the desired goal of status epilepticus even in a single animal despite repeated attempts with the standard dosage. With modified pilocarpine dosage (as described above), we could achieve a 58% seizure induction rate while maintaining lower mortality rates. MTLE model creation was validated by histological examination of the hippocampus. In the current report, we discuss the difficulties encountered during the generation of this model, how troubleshooting was done, and propose a protocol based on these findings.

Animal Selection and Housekeeping

A. Age and Weight: SE was not observed in heavy rats weighing 350-400 grams even with the modified dosage; however, small Wistar rats weighing 100-150 grams showed SE generation. The increased occurrence of seizures in small rats as compared to large rats was in accordance with those of Hirsch et al., 1992 [43] and Curia et al., 2008 study [44], in which rats of P11-30 (P: postnatal) were taken, SE was induced in all small rats, and the mortality rate varied from zero% (in P11-14) to an intermediate 33% (in P15-21) spiking up to 50% (in P22-30), signifying that the mortality rate increased with the age and weight of rats. The chemicals used in the generation of the LPISE model are weight-dependent; an increased dose of pilocarpine required for seizure induction in heavy rats may lead to severe toxicity and death [45, 46].

B. Sex: Only male rats were used as female rats are more resistant to SE induced by pilocarpine due to estradiol causing relative

insensitivity to cholinergic drugs. In females, the different phases of the estrous cycle show differences in the efficacy of pilocarpine at the estradiol peak of the estrus cycle [47, 48]. Lawrence et al., 2010 reported that progesterone-derived neurosteroids (e.g., pregnanolone, allopregnanolone) affect spontaneous recurrent seizures (SRS) in female rats injected with pilocarpine [49].

C. Feed and Bedding: In an attempt to normalize all physiological and environmental conditions, AIN-76 rodent diet was used as a standard reference diet to reduce feed variability [50], and corncob bedding was provided to the animals because it inhibits the accumulation of ammonia. Good husbandry practices of changing bedding frequently, sanitation, and frequent disposal of feces were religiously followed.

Model Creation Mechanism

It has been documented that the administration of LiCl 18-22 hours prior to the injection of pilocarpine and methyl scopolamine before pilocarpine administration successfully induces SE in Wistar rats [36, 37, 42]. Lithium has been shown to potentiate the effect of pilocarpine, thereby decreasing the dose of pilocarpine by 10 times, ultimately reducing the mortality rate of rats to a greater extent.

Lithium, when administered to rats prior to pilocarpine, increases acetylcholine release in the hippocampus and results in more acetylcholine crossing the synaptic cleft and reaching the postsynaptic membrane, where it activates M1 muscarinic receptors and further decreases the dose of pilocarpine by indirectly activating T-lymphocytes and mononuclear cells by complementing the pilocarpine action [51-53]. Pilocarpine induces peripheral inflammation, enhancing blood-brain barrier (BBB) permeability, which further causes its diffusion in the brain resulting in an imbalance between excitatory and inhibitory transmission causing SE [54-56].

Pilocarpine is a muscarinic receptor (M1) agonist, thus stimulating continuous excitatory activity; this enhanced excitability leads to seizures that build up into a limbic SE [57, 58]. Methyl scopolamine does not cross the BBB and reduces the peripheral effects caused by pilocarpine [59]. The absence of methyl scopolamine results in animals displaying symptoms of peripheral cholinergic activity such as tremor, salivation, piloerection, chromodacryorrhea, and diarrhea after pilocarpine administration [60].

Structural damages and subsequent development of spontaneous recurrent seizures resemble those of human seizures. Moreover, the Li-Pi model is a good mimic of human TLE and is best suited for molecular as well as histological studies because the seizures originate from the mesial temporal

structures of the hippocampal formation, and electrical/chemical changes in brain tissue have been found to be similar to human epilepsy, and pathological changes (mesial temporal lobe sclerosis) are comparable to human pathology [14].

Mortality: When pilocarpine is administered in a single dose (30mg/kg), then mortality is reported to be 90-92% [33]. But when the same dose is split (10mg/kg each), then mortality is reduced by 50% [61]. In our present modified protocol, we have evidenced a 10% mortality. Dehydration and exhaustion are considered to be the main reasons for mortality. Diazepam helps by arresting the SE. Nutritional support in the form of regular feeding of glucose by oral gavage and moistening the dry eyes were some of the practices which we found useful.

Possible Reasons for Resistance to Model Development:

During this study, the inability to reproduce the LPISE model with the standard recommended protocol as per the literature is difficult to explain because all the factors with the potential to create variability were judiciously monitored, and multiple attempts were taken. Prior to this report, the lack of reproducibility of the LPISE model in different labs has already been documented [62-64]. Pilocarpine-resistant animals have been cited as the cause of difficulty in seizure induction.

Pilocarpine resistance could be due to a particular rodent strain, species, or gender. This variability could also be attributed to the increased expression of drug efflux transporters on the BBB. Efflux transporters have been extensively studied in relation to drug resistance in epilepsy. The main efflux transporters are Permeability glycoprotein (P-gP) and Multiresistance protein-1 (MRP-1). Their physiological role is to extrude harmful substances at the blood-brain barrier (BBB) so that the internal milieu of the brain is preserved. They have been found to be overexpressed in drug-resistant epileptic patients leading to subclinical concentrations of antiepileptic drugs at the epileptic focus.

Similarly, efflux transporters may extrude the pilocarpine and consequently reduce the bioavailability of the drug at the target site [65]. The saturation level of muscarinic receptors causing resistance of rats to further increased doses of pilocarpine might result in this failure. When the agonist concentration is high, the receptors get saturated, which may lead to receptor downregulation and ultimately lead to resistance. Thus, there is a need of standardization of protocol to develop LPISE model, considering practical problems, so that the research on translational preclinical epilepsy is comparable [62, 66-69].

Proposed method of model development:

Young small-sized rats (100-150g) should be chosen. To ensure the drug reaches the

peritoneum, we recommend using a 26G needle, which is longer (38mm) than the routinely used 24G needle. LiCl (127mg/kg, i.p.) should be injected, followed by methyl scopolamine (1mg/kg, i.p.) after 18-22 hours. Precisely 30 minutes later, injection of pilocarpine via the intraperitoneal route is recommended, starting with incremental doses from a minimum of 30 mg/kg, up to a maximum total dose of 90 mg/kg of pilocarpine. Once the rats begin to develop seizures (usually after an interval of 40-50 minutes), diazepam (10mg/kg) should be injected to reduce seizure severity and increase survival rate.

Since the rats are fatigued due to seizures and unable to eat and drink on their own, glucose (10% of body weight) should be administered to rats through oral gavage as an energy drink to endure the severity of seizures. Rats that did not develop seizures should undergo a wash-out period of 48 hours and can then be reused for model generation; this aligns with the ethics of using the minimal number of animals. It is noteworthy that a mortality rate of 10% was observed during our model generation.

CONCLUSION

The current paper highlights the various adjustments made to the original lithium pilocarpine model to address both the high mortality and low induction rates. These modifications led to improved seizure induction rates (58%) and lower mortality rates (10.52%). The protocol proposed by us meets all three criteria laid down for an ideal animal model of epilepsy [12, 13]. It demonstrates constructive validity, as evidenced by the typical histological features observed in the hippocampal tissue. Furthermore, typical stages of epilepsy were demonstrated, representing face validity, and we have observed predictive validity as it responds to the anti-epileptic drug valproic acid (Results not shared in this paper).

Limitations of the present study:

Since we intend to study acute events such as changes in calcium levels, neuroinflammation in tissue samples, oxidative stress-related changes, etc., the model development was conducted, and rats were sacrificed after 24 hours for downstream processes like histology, gene, and protein expression. The time frame of 24 hours appears to be a potential limitation for the study of complications related to model development, as longer-term issues are not addressed.

REFERENCES

1. Trinka E, et al. A definition and classification of status epilepticus–Report of the ILAE Task Force on Classification of Status Epilepticus. Epilepsia. 2015;56(10):1515-1523.
2. Srinivas H, Shah U. Comorbidities of epilepsy. Neurol India. 2017;65(7):18.
3. Laxer KD, et al. The consequences of

refractory epilepsy and its treatment. Epilepsy Behav. 2014;37:59-70.
4. Löscher W, et al. Drug resistance in epilepsy: clinical impact, potential mechanisms, and new innovative treatment options. Pharmacol Rev. 2020;72(3):606-638.
5. Mohanraj R, et al. Mortality in adults with newly diagnosed and chronic epilepsy: a retrospective comparative study. Lancet Neurol. 2006;5(6):481-487.
6. Trinka E, et al. Cause-specific mortality among patients with epilepsy: results from a 30-year cohort study. Epilepsia. 2013;54(3):495-501.
7. Ethemoğlu ÖU, et al. The Relationship Between Clinico-Pathological Properties and p-Glycoprotein Expression in Hippocampal Sclerosis Among Patients with Mesial Temporal Lobe Epilepsy Who Undergo Selective Amygdalohippocampectomy Operation. Arch Neuropsychiatry. 2020;57(3):204.
8. Weidner LD, et al. The expression of inflammatory markers and their potential influence on efflux transporters in drug-resistant mesial temporal lobe epilepsy tissue. Epilepsia. 2018;59(8):1507-1517.
9. Aronica E, et al. Expression and cellular distribution of multidrug resistance–related proteins in the hippocampus of patients with mesial temporal lobe epilepsy. Epilepsia. 2004;45(5):441-451.
10. Lachos J, et al. Characterization of the gene expression profile of human hippocampus in mesial temporal lobe epilepsy with hippocampal sclerosis. Epilepsy Res Treat. 2011;2011.
11. Kollias G, et al. Animal models for arthritis: innovative tools for prevention and treatment. Ann Rheum Dis. 2011;70(8):1357-1362.
12. Willner P. Animal models as simulations of depression. Trends Pharmacol Sci. 1991;12:131-136.
13. van der Staay FJ. Animal models of behavioral dysfunctions: basic concepts and classifications, and an evaluation strategy. Brain Res Rev. 2006;52(1):131-159.
14. Kandratavicius L, et al. Animal models of epilepsy: use and limitations. Neuropsychiatr Dis Treat. 2014;10:1693-1705.
15. Dhir A. Curcumin in epilepsy disorders. Phytother Res. 2018;32(10):1865-1875.
16. Lévesque M, Avoli M, Bernard C. Animal models of temporal lobe epilepsy following systemic chemoconvulsant administration. J Neurosci Methods. 2016;260:45-52.
17. Kienzler-Norwood F, et al. A novel animal model of acquired human temporal lobe epilepsy based on the simultaneous administration of kainic acid and lorazepam. Epilepsia. 2017;58(2):222-230.

18. Ratté S, Lacaille J-C. Selective degeneration and synaptic reorganization of hippocampal interneurons in a chronic model of temporal lobe epilepsy. Adv Neurol. 2006;97:69-76.
19. Gröticke I, Hoffmann K, Löscher W. Behavioral alterations in a mouse model of temporal lobe epilepsy induced by intrahippocampal injection of kainate. Exp Neurol. 2008;213(1):71-83.
20. Reddy DS, Kuruba R. Experimental models of status epilepticus and neuronal injury for evaluation of therapeutic interventions. Int J Mol Sci. 2013;14(9):18284-18318.
21. Cronin J, Dudek FE. Chronic seizures and collateral sprouting of dentate mossy fibers after kainic acid treatment in rats. Brain Res. 1988;474(1):181-184.
22. Cavalheiro E, Riche D, La Salle GLG. Long-term effects of intrahippocampal kainic acid injection in rats: a method for inducing spontaneous recurrent seizures. Electroencephalogr Clin Neurophysiol. 1982;53(6):581-589.
23. Minjarez B, et al. Behavioral changes in models of chemoconvulsant-induced epilepsy: A review. Neurosci Biobehav Rev. 2017;83:373-380.
24. Pitkänen A, Galanopoulou AS, Moshé SL. Models of seizures and epilepsy. Academic Press; 2017.
25. Cavalheiro E, et al. Long-term effects of pilocarpine in rats: structural damage of the brain triggers kindling and spontaneous I recurrent seizures. Epilepsia. 1991;32(6):778-782.
26. Mello LE, Cavalheiro EA, Tan AM, Kupfer WR, Pretorius JK, Babb TL, Finch DM. Circuit mechanisms of seizures in the pilocarpine model of chronic epilepsy: cell loss and mossy fiber sprouting. Epilepsia. 1993 Dec;34(6):985-95.
27. Blümcke I, Thom M, Aronica E, Armstrong DD, Vinters HV, Palmini A, Jacques TS, Avanzini G, Barkovich AJ, Battaglia G, Becker A, et al. Defining clinico-neuropathological subtypes of mesial temporal lobe epilepsy with hippocampal sclerosis. Brain Pathol. 2012 May;22(3):402-11.
28. Mello LG, Mello LE, Cavalheiro EA. Granule cell dispersion in relation to mossy fiber sprouting, hippocampal cell loss, silent period and seizure frequency in the pilocarpine model of epilepsy. Epilepsy Res Suppl. 1992;9:51-9; discussion 59.
29. Isokawa M, Mello LE. NMDA receptor-mediated excitability in dendritically deformed dentate granule cells in pilocarpine-treated rats. Neurosci Lett. 1991 Dec 16;129(1):69-73.
30. Fujikawa DG, Shinmei SS, Cai B. Lithium-pilocarpine-induced status epilepticus produces necrotic neurons with

internucleosomal DNA fragmentation in adult rats. Eur J Neurosci. 1999 May;11(5):1605-14.

31. Glien M, Brandt C, Potschka H, Löscher W. Repeated low-dose treatment of rats with pilocarpine: low mortality but high proportion of rats developing epilepsy. Epilepsy Res. 2001 Mar;46(2):111-19.
32. Jope RS, Morrisett RA, Snead OC 3rd. Characterization of lithium potentiation of pilocarpine-induced status epilepticus in rats. Exp Neurol. 1986 Dec;91(3):471-80.
33. Morrisett RA, Jope RS, Snead OC 3rd. Effects of drugs on the initiation and maintenance of status epilepticus induced by administration of pilocarpine to lithium-pretreated rats. Exp Neurol. 1987 Jan;97(1):193-200.
34. Modebadze T, Morgan NH, Perucca P, Moshé SL, Löscher W. A low mortality, high morbidity reduced intensity status epilepticus (RISE) model of epilepsy and epileptogenesis in the rat. PLoS One. 2016 Feb 11;11(2):e0147265.
35. Needs HI, Rikani AA, Benesova J, Sisodiya SM, Thom M, Gorter JA, Potschka H, Galanopoulou AS, Kokaia M. Changes in excitatory and inhibitory receptor expression and network activity during induction and establishment of epilepsy in the rat Reduced Intensity Status Epilepticus (RISE) model. Neuropharmacology. 2019 Mar;158:107728.
36. Faure JB, Akimana G, Amorim IS, Nunes B, Costenla AR, Neves-Carvalho A, et al. A comprehensive behavioral evaluation in the lithium-pilocarpine model in rats: effects of carisbamate administration during status epilepticus. Epilepsia. 2013 Jul;54(7):1203-13.
37. Wang P, Liu X, Cao L, Zhang H, Shao Q, Wang B, et al. Predicting signaling pathways regulating demyelination in a rat model of lithium-pilocarpine-induced acute epilepsy: A proteomics study. Int J Biol Macromol. 2021 Mar 1;193:1457-70.
38. Xiang T, Chen X, He Z, Liu Y, Zhao D, Wu H, et al. Klotho ameliorated cognitive deficits in a temporal lobe epilepsy rat model by inhibiting ferroptosis. Brain Res. 2021 Jan 1;1772:147668.
39. Salem NA, Radwan NM, Ahmed HH, Khadrawy YA, Aboul Ezz HS, El-Shamarka ME. New prospects of mesenchymal stem cells for ameliorating temporal lobe epilepsy. Inflammopharmacology. 2018 Oct;26(5):963-72.
40. Bojja SL, Kasi B, Reddy YR, Mathala V, Kandadai RM, Digumarti RR, et al. Metformin ameliorates the status epilepticus-induced hippocampal pathology through possible mTOR modulation. Inflammopharmacology. 2021 Feb;29(1):137-51.
41. Hui L, Ning W, Nan S, Shulan Z, Jianguo J, Baolu Z. The neuroprotection of insulin on ischemic brain injury in rat

hippocampus through negative regulation of JNK signaling pathway by PI3K/Akt activation. Brain Res. 2005 Jun 1;1052(1):1-9.

42. Fan J, Ma H, Ma L, Fan X, Sun J, Wang X, et al. Neural activities in multiple rat brain regions in lithium-pilocarpine-induced status epilepticus model. Front Mol Neurosci. 2020 Jul 28;13:323.
43. Hirsch E, Baram T, Snead OC 3rd. Ontogenic study of lithium-pilocarpine-induced status epilepticus in rats. Brain Res. 1992 Mar 6;583(1-2):120-26.
44. Curia G, Lucchi C, Vinet J, Gualtieri F, Marinelli C, Torsello A, et al. The pilocarpine model of temporal lobe epilepsy. J Neurosci Methods. 2008 Oct 15;172(2):143-57.
45. Esclapez M, Hirsch JC, Ben-Ari Y, Bernard C. Newly formed excitatory pathways provide a substrate for hyperexcitability in experimental temporal lobe epilepsy. J Comp Neurol. 1999 Nov 15;408(4):449-60.
46. Goffin K, Nissinen J, Van Laere K, Pitkänen A. Cyclicity of spontaneous recurrent seizures in pilocarpine model of temporal lobe epilepsy in rat. Exp Neurol. 2007 Oct;205(2):501-5.
47. Scharfman HE, MacLusky NJ. Sex differences in the neurobiology of epilepsy: a preclinical perspective. Neurobiol Dis. 2014 Nov;72 Pt B:180-92.
48. Zuo D, Wang L, Wang X, Xu J, Yang Y, Wang G, et al. The novel estrogen receptor GPER1 decreases epilepsy severity and susceptivity in the hippocampus after status epilepticus. Neurosci Lett. 2020 Oct;728:134978.
49. Lawrence C, Martin BS, Sun C, Williamson J, Kapur J. Endogenous neurosteroid synthesis modulates seizure frequency. Ann Neurol. 2010 May;67(5):689-93.
50. Hogan MC, Norton JN, Reynolds RP. Environmental factors: macroenvironment versus microenvironment. In: Weichbrod RH, Thompson GA, Norton JN, editors. Management of Animal Care and Use Programs in Research, Education, and Testing. CRC Press; 2017. p. 461-77.
51. Hillert MH, Imfeld KL, Neher JJ, van de Berg WD, Riek-Burchardt M, Pagenstecher A, et al. Dynamics of hippocampal acetylcholine release during lithium-pilocarpine-induced status epilepticus in rats. J Neurochem. 2014 Oct;131(1):42-52.
52. Marchi N, Granata T, Janigro D. Antagonism of peripheral inflammation reduces the severity of status epilepticus. Neurobiol Dis. 2009 Aug;33(2):171-81.
53. Duvall A, Gallicchio V. Lithium treatment in clinical medicine: history, current status and future use. J Cell Sci Ther. 2017;8(270):2.

54. Priel MR, Albuquerque E. Short-term effects of pilocarpine on rat hippocampal neurons in culture. Epilepsia. 2002;43:40-6.
55. Marchi N, Oby E, Batra A, Uva L, De Curtis M, Hernandez N, et al. In vivo and in vitro effects of pilocarpine: relevance to ictogenesis. Epilepsia. 2007 Oct;48(10):1934-46.
56. Uva L, Breschi GL, Papouin T, Zonta M, Rossi D. Acute induction of epileptiform discharges by pilocarpine in the in vitro isolated guinea-pig brain requires enhancement of blood-brain barrier permeability. Neuroscience. 2008 Jun 12;151(1):303-12.
57. De Almeida Furtado M, Rossetti F, de Oliveira JA, Scorza FA, Cavalheiro EA, Arida RM, et al. Behavioral, morphologic, and electroencephalographic evaluation of seizures induced by intrahippocampal microinjection of pilocarpine. Epilepsia. 2002;43:37-9.
58. Turski WA, Cavalheiro EA, Schwarz M, Czuczwar SJ, Kleinrok Z, Turski L. Limbic seizures produced by pilocarpine in rats: behavioural, electroencephalographic and neuropathological study. Behav Brain Res. 1983 Jun;9(3):315-35.
59. Davis KL. Brain acetylcholine and neuropsychiatric disease. Springer Science & Business Media; 2013.
60. Vezzani A. Pilocarpine-induced seizures revisited: what does the model mimic? Epilepsy Curr. 2009 Sep-Oct;9(5):146-8.
61. Juvale IIA, Has ATC. The evolution of the pilocarpine animal model of status epilepticus. Heliyon. 2020 Jul;6(7).
62. Dingledine R, Varvel NH, Carlson GC, Tu W. Transcriptional profile of hippocampal dentate granule cells in four rat epilepsy models. Sci Data. 2017 Nov 28;4(1):170061.
63. Landi M, Everitt J, Berridge B. Bioethical, reproducibility, and translational challenges of animal models. ILAR J. 2021;62(1-2):60-5.
64. National Academies of Sciences, Engineering, and Medicine. Advancing disease modeling in animal-based research in support of precision medicine: proceedings of a workshop. National Academies Press; 2018.
65. Kaur M, Krishnan P, Ghosh S, Prasad K, Sharma S. Expressional Study of Permeability glycoprotein (P-gp) and Multidrug Resistance Protein 1 (MRP-1) in Drug-Resistant Mesial Temporal Lobe Epilepsy. Basic Clin Neurosci. 2023 Sep-Oct;14(5):0-0.
66. Gorter JA, van Vliet EA, Rauwé WM, Lopes da Silva FH, Wadman WJ. A companion to the preclinical common data elements for physiologic data in rodent epilepsy models. A report of the TASK3 Physiology Working Group of the

ILAE/AES Joint Translational Task Force. Epilepsia Open. 2018;3:69-89.

67. Krop EJ, Doekes G, Stone MJ, Aalberse RC, Van Der Zee JS. Spreading of occupational allergens: laboratory animal allergens on hair-covering caps and in mattress dust of laboratory animal workers. Occup Environ Med. 2007 Apr;64(4):267-72.
68. Kaur, M., et al., Expressional Study of Permeability glycoprotein (P-gp) and Multidrug Resistance Protein 1 (MRP-1) in Drug-Resistant Mesial Temporal Lobe Epilepsy. Basic and Clinical Neuroscience: p. 0-0.
69. Stein HA, Stein RM, Freeman MI. The Ophthalmic Assistant E-Book: A Text for Allied and Associated Ophthalmic Personnel. Elsevier Health Sciences; 2017.

[Video Link: https://drive.google.com/file/d/1_HZWxCwwj1Utp5-p2A86L1bdgwS6H-w0/view?usp=sharing]

https://doi.org/10.46351/jas.v31i2pp82-89

JAS Journal of Anatomical Sciences (U.P. Chapter of Anatomical Society of India)

Original Article

CORRELATION OF CONSANGUINITY WITH PREVALENCE OF CHROMOSOMAL ANOMALIES IN PATIENTS OF AMBIGUOUS GENITALIA: A CYTOGENETIC STUDY

Anushree Pandey[1], Rakesh Kumar Verma[2], Dewanshi Mishra[3], Archana Rani[2], Navneet Kumar[2]

1. Maharshi Vishwamitra Autonomous State Medical College Ghazipur, India
2. Department of Anatomy, KGMU, Lucknow, India
3. Dr Ram Manohar Lohia Institute of Medical Sciences, Lucknow, India

ABSTRACT

Introduction: The condition when the sex of the baby can't be determined and there is variation of genitalia from normal is known as ambiguous genitalia. Consanguinity can be defined as blood relationship that exists among individuals that descend from a common ancestor. If two individuals who are in close blood relation get married there is high chance that any single copy of gene which is present in common ancestor gets doubled in the subsequent generation. A recessive gene may thus come to light for the first time in subsequent generation. The aim of the present study was to see the correlation of consanguinity with prevalence of chromosomal anomalies in patients of ambiguous genitalia.

Materials and Methods: Study was conducted in the cytogenetic laboratory of the Department of Anatomy, King George's Medical University UP, Lucknow. The patients were screened in the Department of Paediatrics and Paediatric Surgery and blood samples were taken. Cytogenetic analysis was done.

Results: Consanguinity was traced in 4 (18.2%) cases, of whom 3 (75%) had chromosomal anomalies. Out of remaining 18 (81.2%) cases, chromosomal anomalies were seen in 4 (22.2%) cases.

Conclusions: The proportion of cases with anomalies was higher in those positive for consanguinity as compared to those without consanguinity.

Keywords : Consanguinity, Ambiguous genitalia, Chromosomal anomalies

Address for Correspondence:
Dr. Anushree Pandey, Assistant Professor, Department of Anatomy, Maharshi Vishwamitra Autonomous State Medical College, Ghazipur, India
Email: anu031103@gmail.com Mob: 8318660751

Date of Receiving: 27 Aug 2023
Date of Acceptance: 30 Sep 2023
ISSN: 0970-1842

INTRODUCTION

Consanguinity is a very common cultural practice socially accepted by nearly 20% of the world's population living in Afro-Eurasia and by people migrating from these regions to other areas. These populations have their own reasons to promote consanguineous marriages. However, there are many drawbacks associated with these marriages, such as higher fertility rates, stillbirths, and slightly elevated infant mortality rates, along with a birth defect frequency that is around 2-3% higher. Autosomal recessive disorders become more apparent in individuals who are offspring of closely blood-related parents, but the offspring of such parents don't show any variation in the frequency of X-linked recessive disorders or autosomal dominant disorders [1, 2, 3, 4]. Therefore, consanguineous marriages are a matter of concern for all geneticists.

When the marriage is between second cousins or closer, with an inbreeding coefficient (F) of ≥0.0156, it is interpreted as a consanguineous marriage [6]. Here, (F) represents a measure of the proportion of loci at which the offspring of a consanguineous union is expected to inherit identical gene copies from both parents. The inbreeding coefficient is even higher when marriage occurs between double first cousins or uncle-niece pairs [3].

Due to the effect of consanguinity, alleles with abnormalities are concentrated in society because closely blood-related individuals have many alleles that are more or less similar, and if mating occurs, the chances of defective alleles in offspring are enhanced, leading to increased morbidity and mortality due to various genetic diseases. Consanguinity produces many ailments, one of which is ambiguous genitalia.

Previously, various terms were used to define this variation of genitalia from normal, such as intersex, hermaphrodite, pseudo-hermaphrodite, etc., but this was very disappointing to many families [7, 8]. Therefore, it was decided to use a better term in place of intersex, hermaphroditism, and pseudo-hermaphroditism [9-10]. Experts from various fields gathered in Chicago in 2005 (the Chicago Consensus) to coin better terminology and treatment recommendations for this issue, and they proposed a new and more respectful term: disorders of sex differentiation (DSDs). Now, the term DSD is used in place of various confusing terms.

Diagnosis of these cases is done by various methods, one of which is cytogenetics, an emerging field of science in which chromosomal structures are observed and analyzed, along with their properties and actions during cell division, whether in somatic cells or germ cells, and their roles in mitosis and meiosis. This helps to understand how chromosomes, or specifically, genes, influence the phenotype of an individual.

MATERIAL AND METHODS

The study was of a descriptive type. The review board of King George's Medical University UP, Lucknow, approved it. Ethical clearance was also granted by the Ethical Clearance Board of King George's Medical University UP, Lucknow, with vide letter number 2083/Ethics/R.Cell-17. The study was conducted in the Anatomy Department Cytogenetic Lab in collaboration with the Pediatric Surgery Department of King George's Medical University UP, Lucknow. Screening of patients was performed in the OPD of the Pediatric Surgery Department. Patients with a clinical diagnosis of ambiguous genitalia, made by the pediatrician and pediatric surgeon, were included in the study.

The criteria for inclusion in the study were patients who provided consent. Patients who declined to give consent were excluded from the study. A detailed history of patients was obtained, considering various factors that influence the development of Disorders of Sex Differentiation (DSD), and samples of suspected cases were collected from there. Peripheral blood samples were taken, and the samples were analyzed in the cytogenetic laboratory; a karyogram was prepared, and evaluation was conducted..

RESULTS

A total of 24 children with suspected ambiguous genitalia lying in sampling frame were included in the study to solve our

Type	No. of cases	Percentage
Consanguinity	4	16.7
No consanguinity	20	83.3

Table 1. Incidence of consanguinity in study population (n=24)

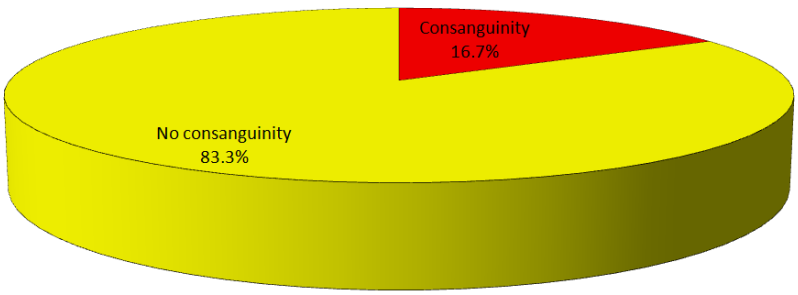


Fig. 1. Pie diagram representing incidence of consanguinity in study population (n=24)

purpose. After history taking and karyotyping following results were found.

Consanguinity was found in four (18.2%) cases, of whom three (75%) had some sort of anomaly in their chromosomes. Among the rest of the 18 (81.2%) cases, a chromosomal anomaly was found in four (22.2%) cases (Table 1 & 2, Fig. 1 & 2). Though the proportion of those with anomalies was higher in those positive for consanguinity compared to those without consanguinity, this difference was not statistically significant (p=0.077). A total of 20 (83.3%) cases did not involve consanguinity. A consanguineous relationship was reported in 4 (16.7%) cases. Among the 4 consanguineous cases, 3 (75%) had different genotypes and phenotypes, while 1 (25%) had the same genotype and phenotype. There were no structural or chromosomal anomalies found among the consanguineous cases (Table 3, Fig. 3).

The occurrence of different genotypes and phenotypes was relatively higher (75%) in consanguineous cases compared to non-consanguineous cases (20%). However, this was not statistically significant (Table 4, Fig. 4).

Consanguinity	**Total**	**With anomalies (n=7)**		**Without anomalies (n=15)**	
		No.	**%**	**No.**	**%**
Yes	4	3	75.0	1	25.0
No	18	4	22.2	14	77.8
		p=0.077 (Fisher exact test)			

Table 2. Association between prevalence of chromosomal anomalies and consanguinity (n=22)

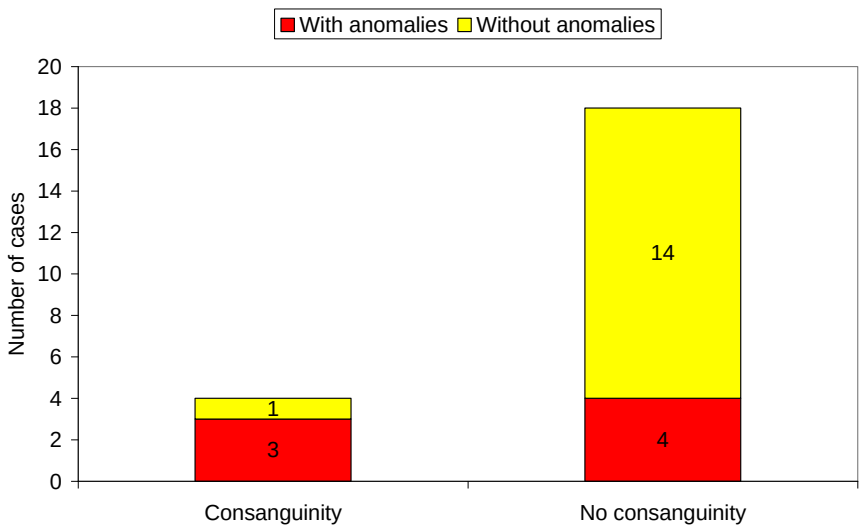


Fig. 2. Bar diagram depicting association between prevalence of chromosomal anomalies and consanguinity

Type	No. of cases	Percentage (%)
Same genotype & phenotype	1	25
Different genotype & phenotype	3	75
Numerical anomalies	0	0
Structural Anomalies	0	0

Table 3. Distribution of anomalies in consanguineous cases (n=4)

Type	Consanguineous cases		Non-consanguineous cases	
	Number	Percentage	Number	Percentage
Same genotype & phenotype	1	25	14	70
Different genotype & phenotype	3	75	4	20
Numerical anomalies	0	0	1	5
Structural Anomalies	0	0	1	5

Table 4. Correlation of types of anomalies found in consanguineous and non-consanguineous cases (n=24)

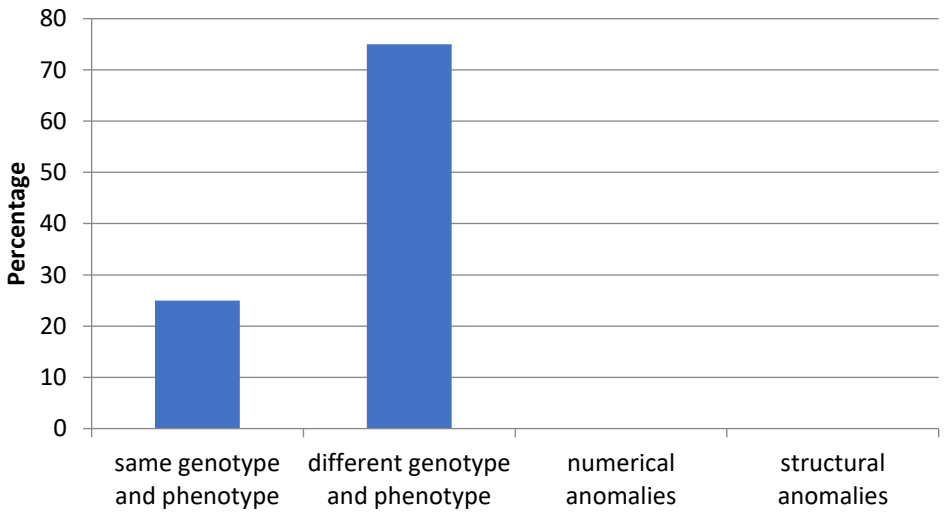


Fig. 3. Bar diagram showing distribution of anomalies in consanguineous cases (n=4)

DISCUSSION

The actual effect of consanguineous marriage on chromosomal abnormalities is still to be meticulously researched and observed, but for autosomal recessive conditions, it can be said that the risk tends to be higher [11]. Many studies have been conducted in different parts of the world by various researchers to determine the exact correlation between consanguinity and various chromosomal abnormalities. During these studies, it was found that in Western countries where consanguineous marriage is less common, the incidence of true ambiguous genitalia was estimated to be 1:5000 births, while in countries where consanguinity was common, the incidence of ambiguous genitalia was higher. In Egypt, it was found to be 1:3000, and in Saudi Arabia, it was found to be 1:2500 [12].

Among 24 patients, we found 4 (16.7%) cases with consanguinity. Our study runs parallel to many other studies but differs from some others. Al-Mutair et al. (2004) retrospectively reviewed a total of 120 medical records of suspected cases of ambiguous genitalia between 1989 and 1999 in Riyadh, Saudi Arabia. They found consanguinity ranged between 60% and 100% in various types of endocrine and congenital developmental defects [13]. Our study differed from this study possibly due to different geographical areas and sample sizes.

Joshi et al. (2006) reviewed 109 patients presenting with ambiguous genitalia over 10 years (1995-2004) at B.J. Wadia Hospital for Children, Parel, Mumbai, India. They found consanguinity in 27 (24.7%) cases [14]. Our study was not entirely consistent with this study but was nearly close to it; the difference might be due to the large sample size and long duration of their study.

Pandith et al. (2015) carried out a study on 50 cases of ambiguous genitalia in the Jammu Kashmir region and found consanguinity in 10 (20%) cases. The result of our study is nearly similar to theirs [15].

Shojaei et al. (2017) conducted a study on 37 patients in Tehran, Iran, and consanguinity was found in 21% of cases. This may be due to the high rate of consanguineous marriages in the Iranian population [16]. Our study is consistent with the results of this study, showing a similar percentage of consanguinity.

CONCLUSION

Consanguinity was found in 16.7% of cases. Among cases born due to consanguineous marriage, 75% had chromosomal anomalies. On comparison with other studies, we noted that consanguinity is one of the important risk factors for the development of ambiguous genitalia, and the rate of chromosomal anomalies was also high. Further studies with better techniques are

needed, and associated chromosomal anomalies need to be investigated. However, accurate diagnosis is burdensome and a big-budget task, so we need to find ways through which we can easily make the diagnosis, and that too on a low budget.

Apart from monetary problems and ease of diagnosis in these cases of ambiguous genitalia, we have to face many religious, social, and cultural factors. Besides diagnostic issues, we also need to educate society about the effects of consanguineous marriages.

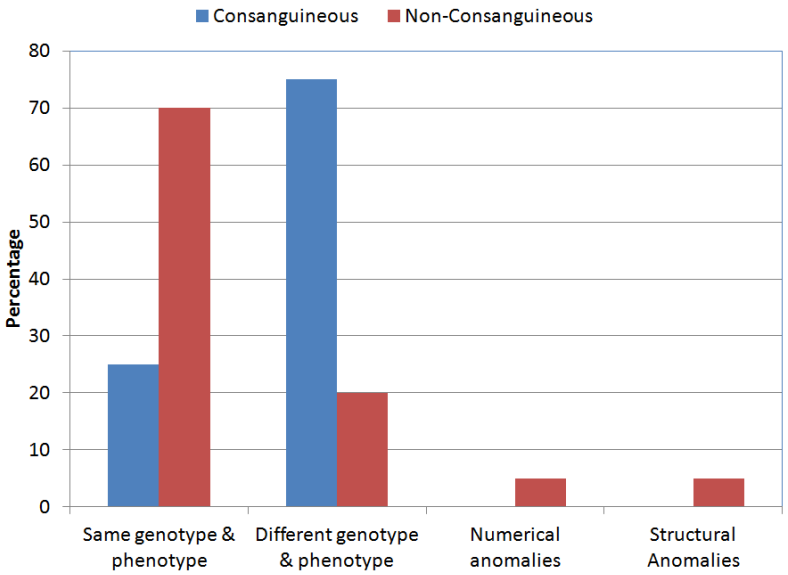


Fig. 4. Bar diagram showing correlation of types of anomalies found in consanguineous and non-consanguineous cases

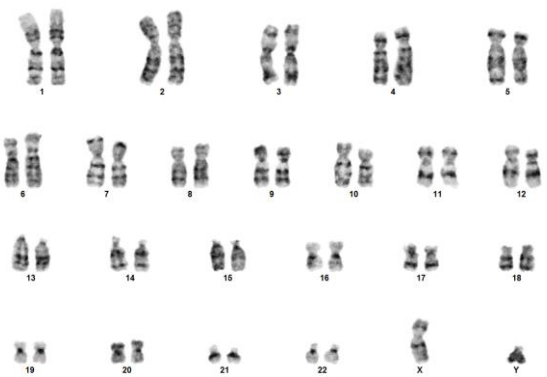


Fig. 5. Karyogram of phenotypic female with male genotype

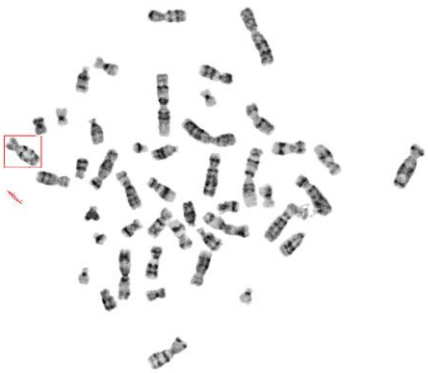


Fig. 6. Metaphase of phenotypic female with male genotype

REFERENCES

1. Bittles AH, Mason WM, Greene J, Rao NA. Reproductive behavior and health in consanguineous marriages. Science. 1991; 252(5007):789-794.
2. Bittles AH, Black ML. The impact of consanguinity on neonatal and infant health. Early Hum Dev. 2010; 86(11):737-741.
3. Hamamy H, Antonarakis SE, Cavalli-Sforza LL et al. Consanguineous marriages, pearls and perils: Geneva International Consanguinity Workshop report. Genet Med. 2011; 13(9):841-847.
4. Tadmouri GO, Nair P, Obeid T, Al Ali MT, Al Khaja N, Hamamy HA. Consanguinity and reproductive health among Arabs. Reprod Health. 2009; 6:17.
5. Hamamy HA, Masri AT, Al-Hadidy AM, Ajlouni KM. Consanguinity and genetic disorders. Profile from Jordan. Saudi Med J. 2007; 28(7):1015-1017.
6. Bittles A. Consanguinity and its relevance to clinical genetics. Clin Genet. 2001; 60(2):89-98.
7. Kim KS, Kim J. Disorders of sex development. Korean J Urol. 2012; 53(1):1-8.
8. Conn J, Gillam L, Conway GS. Revealing the diagnosis of androgen insensitivity syndrome in adulthood. BMJ. 2005; 331(7517):628-630.
9. Hughes IA, Houk C, Ahmed SF, Lee PA. Consensus statement on management of intersex disorders. Arch Dis Child. 2006; 91(7):554-563.
10. Houk CP, Hughes IA, Ahmed SF, Lee PA. Summary of consensus statement on intersex disorders and their management. International Intersex Consensus Conference. Pediatrics. 2006; 118(2):753-757.
11. Amudha S, Aruna N, Rajangam S. Consanguinity and chromosomal abnormality. Indian J Hum Genet. 2005; 11(2):108-110.
12. Bashamboo A, McElreavey K. Consanguinity and disorders of sex development. Hum Hered. 2014; 77(1-4):108-117.
13. Al-Mutair A, Anwar Iqbal M, Sakati N, Ashwal A. Cytogenetics and etiology of ambiguous genitalia in 120 pediatric patients. Ann Saudi Med. 2004; 24(5):368-372.
14. Joshi RR, Rao S, Desai M. Etiology and Clinical profile of ambiguous genitalia - An overview of 10 years experience. Indian Pediatr. 2006; 43:974-979.
15. Pandith AA, Akbar S, Faheem S, Malla TM, Zargar MH, et al. Molecular and cytogenetic evaluation of gender in patients born with ambiguous genitalia from different regions of the valley of Kashmir, North India. J Genet Syndr Gene Ther. 2015; 6(2):1-5.
16. Shojaei A, Ebrahimzadeh-Vesal R, Ali A, Razzaghy-Azar M, Khakpour G, Ghazi F, Tavakkoly-Bazzaz J. Genetic screening of Iranian patients with 46,XY disorders of sex development. Rep Biochem Mol Biol. 2017; 6(1):59-65.

https://doi.org/10.46351/jas.v31i2pp90-99

JAS Journal of Anatomical Sciences (U.P. Chapter of Anatomical Society of India)

Original Article

A ROENTGENOLOGICAL STUDY OF EPIPHYSEAL UNION AROUND LOWER END OF HUMERUS IN BOYS AND GIRLS OF CHITRADURGA REGION OF KARNATAKA

Umesh Choudhary[1], G. M. Mahesh[2], Anand Mishra[1]

1. Department of Anatomy, Institute of Medical Sciences, BHU, Varanasi, India
2. Department of Anatomy, Basaveshwara Medical College and Hospital, Chitradurga, Karnataka, India

ABSTRACT

Introduction: According to previous research on long bones worldwide, there is no consistent sequence for the ossification of long bones in different countries or regions within a single country. The current study was started with the aforementioned considerations in mind, as well as the fact that Chitradurga district has less such work done. In medicine, law, and public health, having exact understanding of the union of the lower end of the humerus will be beneficial.

Materials and Methods: In the Chitradurga area of central Karnataka, India, a radiological investigation of the lower end of the humerus was performed on a total of 97 males and 108 females in the age range of 11 to 20 years. The radiographs were thoroughly examined, and the results were noted.

Results: Fusion of the epiphyseal center of lateral epicondyle with the capitulum was observed between the age group of 13 to 14 years in males and11-12 years in females. Fusion of the Trochlea and capitulum was observed in females between the age group of 11 to 12 years and in males between the age group of 12 to 13 years. In males between the age group of 14 to 17 years and in females between the age group of 11 to 14 years there was fusion of the medial epicondyle with the shaft. In males aged 14 to 15 years and females aged 12 to 13 years fusion of conjoint epiphysis with the shaft was observed.

Conclusions: In comparison to males, females typically experience epiphyseal fusion 1- 3 years earlier.

Keywords : Epicondyle, Ossification, Humerus

Address for Correspondence:
Dr. Umesh Choudhary, Associate Professor, Department of Anatomy, Institute of Medical Sciences, BHU, Varanasi, India
Email: dr.umesh2311@gmail.com Mob: 9717434562

Date of Receiving: 14 Sep 2023
Date of Acceptance: 24 Oct 2023
ISSN: 0970-1842

INTRODUCTION

Bone is essentially a highly vascular, living, constantly changing, and mineralized special connective tissue, remarkable for its characteristic growth mechanism. Bone serves various functions such as support, movement, sound transduction, protection, production, attachment, and storage in the body. The humerus is one of the long bones of the body present in the brachium region, forming the upper arm and joining it to the shoulder and forearm.

A long bone has two ends called Epiphysis and a middle section called Diaphysis. The age period during which bone epiphyses combine is remarkably stable for a specific epiphysis. In a range of medical and surgical disciplines, as well as in the medico-legal arena, it holds significant value. Knowledge of normal ossification is, therefore, important medically, as the size and relation of epiphysis vary with age. According to reports, law courts worldwide recognize the examination of the epiphyseal union of bones as a valid scientific technique for determining an individual's age.

Numerous states in India and overseas have conducted extensive research on the age of epiphyseal unions, and based on the findings of these studies, it is clear that there are variations in epiphyseal union ages not only between Indian states but also between foreign countries.

This difference is thought to be caused by dietary habits, genetics, metabolism, climate, and region. In light of the aforementioned information, this study was carried out to determine the epiphyseal union at the lower end of the humerus to estimate an individual's age. The results were compared to those of regional Indian and Western populations to address the gaps and scarcity of such data in the medical literature.

MATERIAL AND METHODS

In the Chitradurga district of Karnataka, 97 males and 108 females between the age group of 11 to 20 who were enrolled in I) Sri Jagadguru Mallilarjuna Murugharajendra English Medium School, II) Bapuji Public School, III) Sri Jagadguru Mallilarjuna Murugharajendra College of Pharmacy, and IV) Sri Jagadguru Mallilarjuna Murugharajendra Institute of Technology-Chitradurga were the subjects of the current study.Diagnosis of these cases is done by various methods, one of which is cytogenetics, an emerging field of science in which chromosomal structures are observed and analyzed, along with their properties and actions during cell division, whether in somatic cells or germ cells, and their roles in mitosis and meiosis. This helps to understand how chromosomes, or specifically, genes, influence the phenotype of an individual.

Random sampling technique was used to choose study participants. Consent was obtained from parents and students.

A thorough medical history, including all necessary information such as name, age, sex, food preferences, socioeconomic situation, and religion, was recorded in the proforma. Standard techniques were employed to record the subjects' height and weight. The age, with respect to their sex, was divided into 9 groups with a minimum of 10 individuals in each group.

The radiographs were taken using the digital Siemens system at the Radiodiagnosis department. The findings were confirmed by Radiologist Dr. Naveen Kumar. The Das Gupta, Vinod Prasad & Shamer Singh (1971) and Banerjee & Agarwal (1998) method of classification were followed during observation of the radiographs showing both complete and incomplete union at the lower end of the humerus.

RESULTS

In the age group of 11 – 12 years:

In males, the ossification center for the lateral epicondyle had not yet developed in 90% of cases, while it had in 10% of cases. In 100% of instances, the medial epicondyle and conjoint epiphyses were not fused with the shaft. Nine percent of the females in this age range had the medial epicondyle fused with the shaft. The lateral epicondyle, capitulum, and trochlea fused together to produce a conjoint epiphysis in 27.2% of the females; this conjoint epiphysis was not united with the shaft. The ossification center for the lateral epicondyle had not yet developed in 36.3% of the females, while it had in 63.6% of them.

In the age group of 12 – 13 years:

In 100% of the males, the medial epicondyle did not fuse with the shaft. The lateral epicondyle was visible in 70% of instances. In 10% of cases, the lateral epicondyle and capitulum were fused together. In 100% of the cases, the conjoint epiphysis was not fused with the shaft. Among the females, 30% of cases showed the union of the epiphysis of the medial epicondyle with the shaft. The conjoint epiphysis fused with the shaft in 30% of cases.

In the age group of 13 – 14 years:

Among the males in this age group, the epiphysis of the medial epicondyle did not fuse with the shaft in 100% of cases. The lateral epicondyle did not appear in 7.14% of cases. The capitulum fused with the trochlea but not with the shaft in 81.8% of cases. The capitulum fused with the lateral epicondyle in 63.6% of cases. Among the females in this age group, 64.2% of cases showed the epiphysis of the medial epicondyle not fused with the shaft. The conjoint epiphysis was fused to the diaphysis in 78.5% of cases. 35.7% of cases showed complete union with the diaphysis.

In the age group of 14 – 15 years:

Among males in this age group, 90% of cases showed the epiphysis of the medial

Age in years	No. of cases			L.E. + Cap.(not fused to Tro) (Not fused to shaft)		Cap + Tro (not fused to L.E.)(not fused to shaft)		L. E. + Tro + Cap (Not fused to shaft)		C.E. fused to shaft		M.E. fused to shaft		Cap + L.E.(not fused to Tro) fused to shaft		Cap + Tro (not fused to L.E.) fused to shaft	
	M	F	TOTAL	M	F	M	F	M	F	M	F	M	F	M	F	M	F
11-12	10	11	21	0	1	0	8	0	1	0	0	0	1	0	1	0	5
12-13	10	10	20	0	0	5	0	1	2	0	8	0	3	0	0	2	0
13-14	11	14	25	1	0	0	0	6	0	0	13	0	5	0	0	0	1
14-15	10	10	20	0	0	0	0	5	1	5	9	1	9	0	0	0	0
15-16	10	10	20	0	0	0	0	8	0	3	9	3	9	0	0	0	0
16-17	12	10	22	0	0	4	0	3	0	5	10	3	10	0	0	2	0
17-18	11	12	23	0	0	0	0	0	0	11	12	11	12	0	0	0	0
18-19	12	15	27	0	0	0	0	0	0	12	15	12	15	0	0	0	0
19-20	11	16	27	0	0	0	0	0	0	10	17	10	17	0	0	0	0
Grand total	97	108	205														

Table 1. The epiphyseal union at the lower end of humerus

Sl. No.	Epiphysis	Males	Females
1	Cap + L.E.	13 yrs 02 mon 18 days	11 yrs 14 days
2	Cap + Tro	12 yrs 01 mon 01 day	11 yrs 14 days
3	L.E. + Tro + Cap	12 yrs 06 mon 01 day	11 yrs 14 days
4	C.E. to shaft	12 yrs 06 mon 28 day	12 yrs 10 days
5	M.E. to shaft	11 yrs 21 days	11 yrs 04 mon 29 days
6	Cap + L.E. to shaft	12 yrs 06 mon 28 days	11 yrs 14 days
7	Cap + Tro to shaft	12 yrs 06 mon 28 days	11 yrs 04 mon 29 days

Table 2. Earliest age of union of different epiphysis

epicondyle and conjoint epiphysis not fused with the diaphysis. Only 10% of cases showed complete union with the diaphysis. Among the females in this age group, 90% of cases showed complete union of the epiphysis of the medial epicondyle and conjoint epiphysis with the shaft. The epiphysis of the medial epicondyle and conjoint epiphysis were not fused with the shaft in 10% of cases.

In the age group of 15 – 16 years:

Among males in this age group, 20% of cases showed the epiphysis of the medial epicondyle and conjoint epiphysis fused with the diaphysis. The lateral epicondyle, capitulum, and trochlea fused with each other but not with the shaft in 80% of cases; 20% of cases showed complete union with the shaft. Among females in this age group, 100% of cases showed complete union of the epiphysis of the medial epicondyle with the shaft. Also, the conjoint epiphysis showed complete union with the shaft in 100% of cases.

In the age group of 16 – 17 years:

Among males in this age group, 25% of cases showed the epiphysis of the medial epicondyle fused with the diaphysis; the conjoint epiphysis was united to the shaft in 41.66% of cases; 25% of cases showed complete union with the shaft. Among females in this age group, 100% of cases showed complete union of the epiphysis of the medial epicondyle with the shaft. Also, the conjoint epiphysis showed complete union with the shaft in 100% of cases.

Sl. No.	Epiphysis	Males	Females
1	Cap + L.E.	13 yrs 02 mon 22 days	11 yrs 04 mon 12 days
2	Cap + Tro	12 yrs 09 mon	11 yrs 05 mon 06 days
3	L.E. + Tro + Cap	16 yrs 09 mon 19 days	14 yrs 03 mon 19 days
4	C.E. to shaft	16 yrs 08 mon 14 days	14 yrs 02 mon
5	M.E. to shaft	16 yrs 08 mon 14 days	14 yrs 02 mon
6	Cap + L.E. to shaft	16 yrs 08 mon 14 days	11 yrs 04 mon 12 days
7	Cap + Tro to shaft	12 yrs 07 mon 15 days	11 yrs 05 mon 06 days

Table 3. Latest age of non-union of different epiphysis

Workers	Subjects	Trochlea fuses to Capitulum (in yrs)		L.E fuses to Capitulum (in yrs)		Trochlea + Capitulum + Lateral epicondyle (in yrs)		Conjoint epiphysis fuses to shaft (in yrs)		M.E. fuses to shaft (in yrs)	
	Indian	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**
Hepworth(1929)	Punjabis	-	-	14	15	-	-	14	15	14 1/2	14 1/2
Lall and Nat (1934)	Uttar Pradesh (Males)	-	-	-	-	-	-	-	15-17	-	15-17
Pillai (1936)	Madrasis	-	-	13-14	13-14	-	-	13-14	13-14	14-17	14-17
Basu and Basu (1938)	Bengalis (Females)	12-13	-	12-13	-	-	-	12-13	-	13-14	-
Galstaun (1939)	Bengalis	9-13	11-15	10-12	11-16	-	-	-	-	14	16
Lall and Townsend (1939)	Uttar Pradesh (Females)	-	-	-	-	-	-	-	-	14-15	-
Franklin (1962)	Vidarbha M.S. (Females)	13-14	-	13-14	-	-	-	13-14	-	14-15	-
Das Gupta et al (1971)	Uttar Pradesh	-	-	-	-	-	-	-	-	17-18	18-19
Kothari (1974)	Rajasthan	11-12	14-15	11-12	14-15	-	-	14-15	18	14	19-21
Jnanesh (2012)	Karnataka	12-13	15-16	12-13	15-16	-	-	12-13	16-17	14-15	18-19
	Other countries										
Davis and Parsons (1927)	Englanders	-	-	-	16	-	-	-	-	-	20
Patterson (1929)	England	14-15	14-15	14-15	17-18	-	-	14-15	18	14	19-21
Sidhom and Derry (1931)	Egypt	-	-	-	-	-	-	17	17	16-18	-
Flecker (1932)	Australians	-	13	13	13	-	-	-	-	15	16
Present study (2014)		**11-12**	**12-13**	**11-12**	**13-14**	**11-12**	**12-13**	**12-13**	**14-15**	**11-14**	**14-17**

Table 4. Comparative study of epiphyseal union of lower end of the humerus

In the age group of 17 – 18 years:

Among males in this age group, 100% of cases showed the union of the medial epicondyle with the shaft. Additionally, in all instances, the conjoint epiphysis and shaft were found to be united. Among females in this age group, 100% of cases showed complete union of the epiphysis of the medial epicondyle with the shaft. Also, the conjoint epiphysis showed complete union with the shaft in 100% of cases.

In the age group of 18 – 19 years:

Among males, the medial epicondyle and shaft were united in 100% of cases. Additionally, in all instances, the conjoint epiphysis and shaft were found to be united. Among females in this age group, 100% of cases showed complete union of the epiphysis of the medial epicondyle with the shaft. Also, the conjoint epiphysis showed complete union with the shaft in all (100%) of cases.

In the age group of 19 – 20 years:

Among females in this age group, 100% of cases showed the union of the medial epicondyle and conjoint epiphysis with the shaft. In this group, the union of the lower end of the epiphysis, including the epiphysis of the medial epicondyle and conjoint, showed complete union with the shaft in 100% of cases. The epiphyseal union at the lower end of the humerus is shown in table 1. The earliest age of union and the latest age of non-union of different epiphyses are shown in tables 2 & 3, respectively.

DISCUSSION

Appreciable variations in the moment of epiphysis union have been observed by the majority of researchers in this field. The comparative analysis between the results of the study of different workers and the present study are discussed below.

Fusion of trochlea with capitulum:

In the present study, the findings of females in the 11 – 12 years age group are similar to the study by Kothari (Rajasthan), but the fusion occurs 1 – 2 years earlier according to Galstaun (Bengalis) and 1 year later according to the study by Basu and Basu (Bengalis). It was also found to be late by 2 – 3 years in the studies conducted by Patterson (England) and Franklin (Vidarbha) [4-8].

Among males, the present statistical data is close to the studies by previous authors Galstaun, Flecker (Australians), but it is found to be 2 – 3 years earlier compared to the studies of Jnanesh (Karnataka), Kothari, and Patterson.[4, 5, 7, 9, 10].

Fusion of lateral epicondyle with capitulum:

In our study, the lateral epicondyle fuses with the capitulum among females at the age of 11 – 12 years, which is similar to the

findings of Galstaun (Bengalis) and Kothari (Rajasthan). But the studies conducted by Jnanesh (Karnataka) and Basu and Basu (Bengalis) were found to be 1 – 2 years later and also 2 – 3 years late in the studies by Pillai (Madrasis), Hepworth (Punjabis), Franklin (Vidarbha), and Patterson (England) [4-8,11,12]. In males, similar findings are seen by Pillai and Flecker, but the findings of Jnanesh, Hepworth, Kothari, and Patterson were found 1 – 2 years & 2 – 3 years later [4, 7, 9-12].

Fusion of conjoint epiphysis with shaft:

In the present study, fusions in females are similar to those seen in the studies by Jnanesh and Basu and Basu. They are 1 – 2 years later compared to the studies by Franklin and Pillai and 2 – 3 years late compared to the studies by Kothari and Patterson, and 4 years late compared to the studies of Sidhom and Derry (Egypt) [4, 6- 8, 11,13]. In males, it is 1 year late compared to studies by Pillai and 1 – 2 years later compared to studies by Jnanesh, Hepworth, Lall and Nat, Sidhom and Derry, but 3 – 4 years later compared to studies by Kothari and Patterson [4, 7, 10-14].

Fusion of medial epicondyle with shaft:

The present findings are similar to the studies by Basu and Basu, Galstaun, Kothari, and Patterson in females, and 6 months to 1 year later in the studies by Jnanesh, Hepworth, Lall and Townsend, Franklin, and Flecker. It was late by 2 – 3 years in the studies conducted by Das Gupta et al and Sidhom and Derry [4-10,13-16]

In the present study, the medial epicondyle fuses with the shaft in males around 14 – 17 years, which is closely related to the findings of Hepworth, Lall and Nat, Pillai, Galstaun, and Flecker. In some males, the fusion was found 1 – 4 years later compared to the studies conducted by Jnanesh, Das Gupta et al, Kothari, and Davis and Parsons [4,5,9-12, 14, 16, 17].

Taking into consideration the research work done by different workers on epiphyseal union at the lower end of the humerus, a table (Table 4) was drawn.

CONCLUSION

The following conclusions are made after studying the radiographs of 205 selected cases.

I. At the lower end of the humerus:

1. At the ages of 11–12 for females and 13–14 for males, the epiphyseal center of the lateral epicondyle fused with the capitulum.
2. The fusion of the trochlea and capitulum epiphyseal centers was observed in females at 11–12 years and in males at 12–13 years.
3. The fusion of the trochlea, capitulum, and epiphyseal center of the lateral epicondyle to form a compound epiphysis was observed in males at 12–13 years and in females at 11–12 years.

4. In females, the age of fusion of the conjoint epiphysis with the shaft was observed at 12–13 years, while in males, it was observed at 14–15 years.

5. In females, the epiphyseal center of the medial epicondyle fused with the shaft between the ages of 11 and 14 years, and in males between 14 and 17 years.

II. Fusion of epiphysis typically happens in females 1-3 years earlier than in males.

III. Diet had no effect on the fusion. Fig. 1 illustrates the progression of the union of conjoint epiphysis with the shaft in males and females with respect to age. Fig. 2 shows the progression of the union of the medial epicondyle with the shaft in males and females with respect to age.

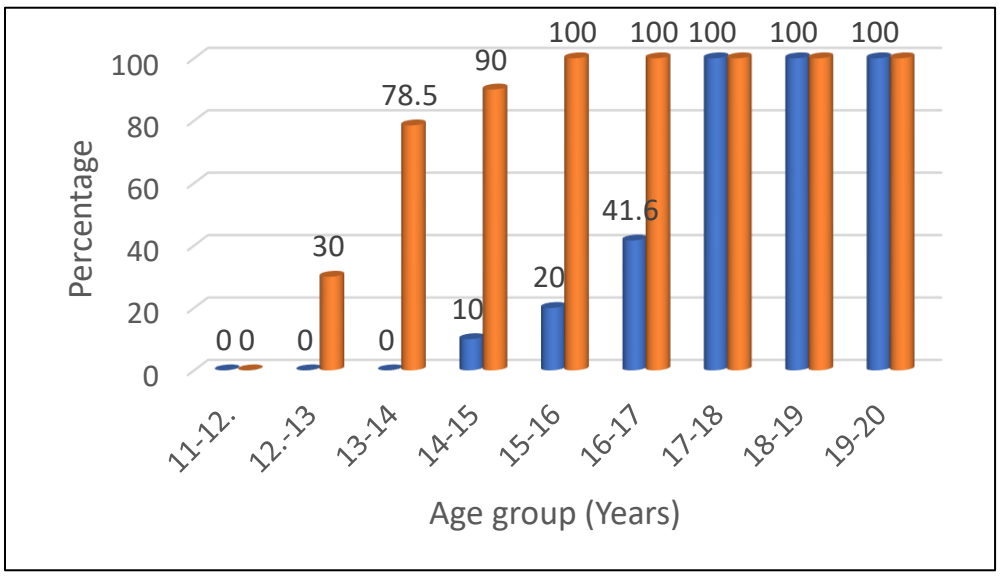


Fig. 1. Progress of union of C.E. With shaft in males and females in relation to age (data fitted in 3-D statistics graph)
Blue: C.E. males; Orange: C.E. females

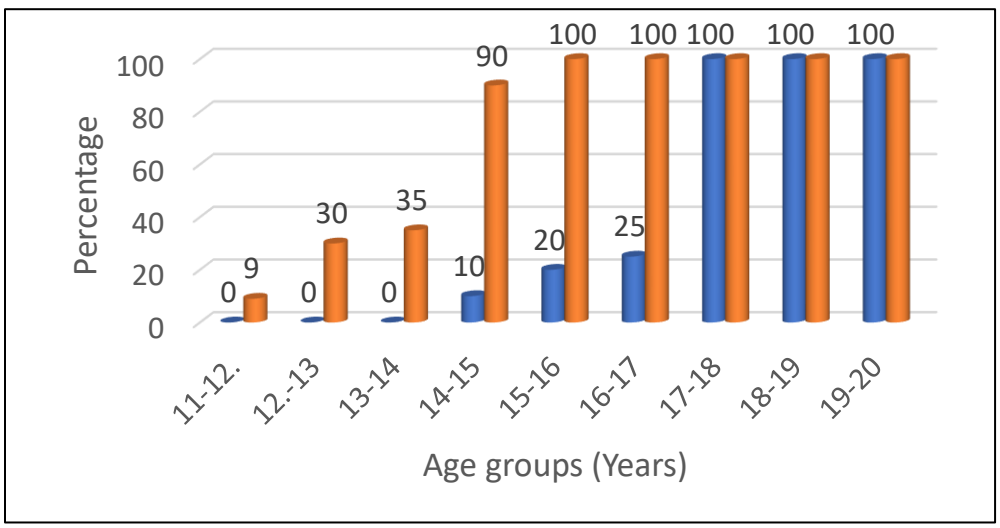


Fig. 2. Progress of union of E.E. with shaft in males and females in relation to age (data fitted in 3-D statistics graph)
Blue: M.E. males; Orange: M.E. females

REFERENCES

1. Standring S, Wigley C. Functional Anatomy of the Muskuloskeletal System. In: Standring S, editor. Gray's Anatomy. 40th ed. China: Churchill Livingstone Elsevier; 2010. p. 85-96.
2. Banerjee KK, Agarwal BB. Estimation of Age from Epiphyseal Union at the Wrist and Ankle Joints in the Capital City of India. Forensic Sci International. 1998;31-39.
3. Choudhary U, Kumar S, Singh A, Bharti P. A radiological study of ossification at the lower end of humerus for age estimation among boys in Central Karnataka, India. Int J Res Med Sci. 2017;5:1204-7.
4. Kothari DR. Age of epiphyseal Union at the Elbow and Wrist Joints in Marwar Region of Rajasthan. J Indian Med Assoc. 1974 Oct;63(8):252-256.
5. Galstaun G. A study of ossification as observed in Indian subjects. Indian J Med Res. 1937 Jul;25(1):267-324.
6. Basu SK, Basu S. A Contribution to the Study of Diaphysioepiphyseal Relation at the Elbow of Young Bengali Girls. Indian J Ped. 1938;5:201-204.
7. Patterson RS. A Radiological investigation of the Epiphysis of the long bones. J Anat. 1929;64:28-46.
8. Franklin CA. A Thesis for M. D. (Forensic Medicine), Radiological study of epiphyseal union in forensic medicine, Lucknow University Lucknow, 1962.
9. Flecker H. Roentgenographic Observation of the Times of Appearance of the Epiphyses and their fusion with the Diaphysis. J Anat Society India. 1932;67:118.
10. Jnanesh RS, Thomas ST, Gowd HS. Estimation of Age by roentgenologic Study of Epiphyseal Union at the Lower end of Humerus in Karnataka. Anatomica Karnataka. 2011;5(1):06-10.
11. Pillai MJS. The Study of Epiphyseal Union for Determining the Age of South Indians. Ind J Med Res. 1936;23:1015-1017.
12. Hepworth SM. On the Determination of Age in Indians from a Study of the Ossification of the Epiphyses of the long bones. Ind Med Gaz. 1929:128.
13. Sidhom G, Derry DE. Dates of Union of some Epiphysis in Egyptians from x-ray photographs. J Anat. 1931;65:196-211.
14. Lall R, Nat BS. Ages of Epiphyseal Union at the Elbow and Wrist Joints amongst Indians. Jr. Ind. Med Res. 1934;21:983-989.
15. Das Gupta SM, Prasad V, Singh S. A Roentgenologic Study of Epiphyseal Union around Elbow, Wrist and Knee Joints and the Pelvis in Boys and Girls of Uttar Pradesh. J Indian Med Assoc. 1974;62:10–12.
16. Lall R, Townsend RS. Ages of Epiphyseal Union at the Elbow and Wrist Joints amongst Indian Girls. Indian Medical Gaette. 1939;74:614-616.
17. Davies DA, Parsons FG. The Age Order of the Appearance and Union of the normal Epiphyses as seen by X-rays. J Anat. 1927;62:58–71.