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Pupil to limbus diameter ratio as an emerging autonomic function test and its correlation with anthropometric parameters in different phases of menstrual cycle

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Abstract

The menstrual cycle is a physiological change in normal females, indicating the proper functioning of both endocrine and reproductive health. The pupil to limbus diameter (PLD) ratio is defined as the ratio of pupil diameter measured at an axial plane to the limbus diameter measured at the same or a parallel axial plane. Alterations in estrogen and progesterone levels may influence cardiac autonomic functions. This study aimed to correlate the PLD ratio with anthropometric parameters, such as body mass index (BMI) and waist circumference (WC), during different phases of the menstrual cycle among medical students.Methods: A cross-sectional study was conducted among first- and second-year female MBBS students aged 17 to 22 years. Anthropometric parameters, including height, weight, BMI, and WC, were measured for all participants. Eye photographs were taken during the follicular and luteal phases of the menstrual cycle, and the diameters were measured manually using the two-box method. The PLD ratios for both eyes showed a significant positive correlation with BMI (right eye: $r = 0.682$, $p < 0.000$; left eye: $r = 0.430$, $p < 0.000$) and waist circumference (right eye: $r = 0.456$, $p < 0.000$; left eye: $r = 0.315$, $p < 0.001$). The PLD ratio can serve as a simple, non-invasive, and cost-effective autonomic function test.

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INTRODUCTION

Menstrual cycle is one among the physiological changes that takes place in a normal female individual, indicating normal functioning of both endocrine and reproductive health [1]. There is sudden withdrawal of hormones estrogen and progesterone resulting in shedding of endometrial lining causing bleeding per vagina every month. Normal days of cycle varies between 21-35 with bleeding for about 3-7 days and volume of blood loss is less than 80 ml. A normal cycle shows intact and functional hypothalamo – pituitary axis. It indicates a balanced state of hormonal and

reproductive system [2]. There are changes in the levels of endogenous sex hormones during different phases of menstrual cycle. There is increase in estrogen during follicular phase which reach its peak prior to ovulation and increase in progesterone during midluteal phase. These alterations may influence cardiac autonomic functions [3].

Autonomic nervous system functioning is important for the maintenance of homeostasis and it is achieved by various mechanisms that regulate blood pressure, body temperature and electrolyte balance [4]. As autonomic functions are associated with multiple system functions, disturbance in the autonomic functions causes dysregulation of the systemic functions. Hence, there is a strong need to evaluate the autonomic functions as a routine test. Literatures have reported a positive correlation between the pupil to limbus diameter ratio and blood pressure and pulse rate [5]. Research studies done on autonomic functions across various phases of menstrual cycle are limited [6].

Ratio of pupil to limbus diameter ratio (PLD) is defined as "the ratio of pupil diameter measured at an axial plane with the limbus diameter measured at a same or parallel axial plane" [7]. PLD ratio is one of the developing autonomic function tests [8]. Pupil is constricted and dilated depending upon the autonomic functions. Pupil dilates with increased sympathetic activity, whereas pupil constricts with increased parasympathetic activity. But the limbus diameter remains constant. So, taking pupil limbus diameter ratio will give an idea about the status of autonomic functions. Understanding the activities of autonomic functions can benefit individuals to begin with various treatment modalities if there are any alterations. However, studies are limited in this area [9].

There are few studies related to autonomic function modulation during different phases of menstrual cycle [6]. However, literatures on pupil to limbus diameter ratio as an autonomic function test in different phases of menstrual cycle and its correlation with anthropometric parameters are not available. Hence, the present study is designed. With this view, the present study was aimed to correlate pupil to limbus diameter ratio with anthropometric parameters like BMI and WC in different phases of menstrual cycle among medical students.

Materials and methods:

The study commenced after obtaining the approval from Institutional Ethics Committee. It was a cross-sectional study conducted among the first year and second year female MBBS students with the age of 17 – 22 years. Voluntary participants of medical students were included for the present study and written informed consent from each of them have been obtained. Information on individual's identity, medical and family history were collected. Anthropometric parameters like height, weight was recorded. Body mass index (BMI) was calculated as weight (kg) divided by height (m^2). Waist circumference (WC) measurement was taken at the end of normal expiration, between the lower rib margin and the iliac crest, with the help of non - elastic measuring tape. Physiological hemodynamic parameters like pulse rate were recorded for one entire minute and blood pressure (BP) has been recorded in participants seated using a Sphygmomanometer [10].

Pupil to limbus diameter ratio (PLD ratio): It was recorded by capturing the photograph of eye using the mobile phone camera. Before capturing the image of the eye, the participants were exposed to the ambient light levels for at least 5 minutes. Luxmeter was used to measure the illumination and it will be kept constant for all the photographs. All the photographs were recorded between 9 am to 11 am in the morning to avoid diurnal variations. After recording the photographs, it was transferred to the power point slides and the diameter was measured manually by the two-box method [11]. The photographs were taken in all the participants during the 2 phases of menstrual cycle namely, follicular phase - 6th day to 13th day of menstrual cycle and luteal phase - 15th day to 28th day or before the next menstrual bleeding [12].

Statistical analysis:

Statistical analyses were performed using SPSS version 21 software. Values were expressed as mean and standard error for the continuous variables. Correlations between the variables were investigated by Pearson's correlation coefficient. Comparison between the groups was analysed by t test. Statistical significance was considered if the p – value is less than 0.05.

Result:

Table 1 Basic characteristics of study population (n = 111) Mean ± SE

Age (Years)	20.32 ± 0.09
BMI (Kg/m2)	21.98 ± 0.32
WC (cm)	74.78 ± 0.70
Pulse (beats/min)	75.18 ± 0.61
SBP (mmHg)	130.93 ± 0.61
DBP (mmHg)	82.99 ± 0.71

Table 2 Mean ± SE of Pupil limbus diameter ratio (PLD ratio) of study population (n = 111)

Variable	Follicular phase	Luteal phase
Pupil limbus ratio of right eye	0.258 ± 0.007	0.230 ± 0.009
Pupil limbus ratio of left eye	0.258 ± 0.007	0.230 ± 0.009

Table 3 Correlation of pupil limbus ratio with body mass index (BMI) and waist circumference (WC) in follicular phase (n = 111)

Parameter		BMI	WC
PLD ratio of right eye in follicular phase	r	0.682	0.456
	p	**0.000***	**0.000***
PLD ratio of left eye in follicular phase	r	0.682	0.456
	p	**0.000***	**0.000***

Table 4 Correlation of pupil limbus ratio with body mass index (BMI) and waist circumference (WC) in luteal phase (n = 111)

Parameter		BMI	WC
PLD ratio of right eye in luteal phase	r	0.430	0.315
	p	**0.000***	**0.001***
PLD ratio of left eye in luteal phase	r	0.430	0.315
	p	**0.000***	**0.001***

*Significant

Basic characteristics of the study population were noted in **Table 1**. The mean age of the participants was 20.32 (±0.09) years. Average pulse was found to be 75.18 ± 0.61. Baseline parameters like body mass index was 21.98 (±0.32) Kg/m^2, average waist circumference was 74.78 (±0.70) cm, mean systolic and diastolic blood pressure were found to be 130.93 (±0.61) and 82.99 (±0.71) mmHg.

In the present study, mean pupil limbus ratio of right eye and left eye in follicular and luteal phase of all the participants were shown in **Table 2** and it was found to be 0.258 ± 0.007 for right and left eye in follicular phase, whereas it was 0.230 ± 0.009 for both the eyes in luteal phase.

Pearson's correlation of pupil limbus ratio of right and left eye in follicular phase with body mass index and waist circumference were depicted in **Table 3**. PLD ratio of right eye and left eye had significant positive correlation (r = 0.682, p <0.000) with BMI and it showed significant positive correlation with waist circumference (r = 0.456, p<0.000). Similarly, Pearson's correlation of pupil limbus ratio of right and left eye in luteal phase with body mass index and waist circumference were depicted in **Table 4**PLD ratio of right eye and left eye had significant positive correlation (r = 0430, p <0.000) with BMI and it showed significant positive correlation with waist circumference (r = 0.315, p<0.001).

Discussion:

The present study was designed to understand the pupil to limbus diameter ratio with anthropometric and physiological hemodynamic parameters in different phases of menstrual cycle among medical students. In this study, volunteers from the medical students were recruited and with them recorded the basic parameters like age, BMI, waist circumference, pulse, systolic and diastolic BP (**Table 1**). Further, pupil limbus ratio was calculated in all the participants by taking photographs of both the eyes during follicular and luteal phases of menstrual cycle (**Table 2**). Ovarian changes seen during the menstrual cycle has follicular, luteal and menstrual phase. These changes are caused by hormonal interactions between hypothalamus, anterior pituitary and ovaries. Granulosa cells in ovarian follicles, corpus luteum and during pregnancy placenta produces estrogen. Corpus luteum formed during luteal phase produces progesterone. Endometrial secretary activity increases because of these hormones. Studies reported protective functions of hormones in luteal phase [13]. Literatures have shown that the female sex hormones have receptors present in cardiovascular system and its expression in heart and also in blood vessels [14].

The present study correlated pupil limbus diameter ratio with body mass index and waist circumference in follicular (**Table 3**) and luteal phases (**Table 4**). There was significant positive

correlation of PLD ratio of both the eyes with BMI and waist circumference in follicular and luteal phases. Diameter of pupil depends on the activity of autonomic nervous system. Dilation of pupil takes place on activation of sympathetic nervous system. But the diameter of limbus is constant [5]. Light entry into the eye is regulated by the pupil and it responds immediately to the external environment changes [15]. Dilator and sphincter pupillae innervated by the sympathetic and parasympathetic branches of autonomic nervous system regulates changes in pupil diameter [16]. During the activation of sympathetic nervous system, dilation of pupil takes place by the release of adrenaline and by parasympathetic inhibition [17]. BMI plays a major role in sympathovagal imbalance because hormones are stored in body fat layer and reduced content of it leads to imbalance in hormones causing disturbance in menstrual cycle producing anovulatory cycle, thereby causing infertility risk. Preventive measures should be employed at an early stage to control weight in an individual so that it can prevent autonomic changes [4].

Conclusion: The present study concludes that the pupil to limbus diameter ratio can be considered as one of the autonomic function tests which is simple, non – invasive and cost – effective. The study will be useful as it reveals the correlation of anthropometric parameters with autonomic functions in different phases of menstrual cycle among medical students, awareness on the importance of autonomic functions can be given to the public.

Ethical Approval

The study commenced after obtaining the approval from Institutional Ethics Committee, with the reference number AIMS/IEC/094/2023 dated 09.09.2023.

Author Contribution

All authors made substantial contributions to the conception, design, acquisition, analysis, or interpretation of data for the work. They were involved in drafting the manuscript or revising it critically for important intellectual content. All authors gave final approval of the version to be published and agreed to be accountable for all aspects of the work, ensuring its accuracy and integrity.

Conflict of Interest

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REFERENCES

[1] Rowland AS, Baird DD, Long S, Wegienka G, Harlow SD, Alavanja M et al, Influence of medical conditions and lifestyle factors on the menstrual cycle. Epidemiology. 13: 668-674, 2002.

[2] Amgain K, Subedi P, Yadav GK, Neupane S, Khadka S and Sapkota SD, Association of Anthropometric Indices with Menstrual Abnormality among Nursing Students of Nepal: A Cross-Sectional Study. Journal of Obesity. 1-8, 2022.

[3] Yazici SYM, Impact of Menstrual Cycle on Cardiac Autonomic Function Assessed by Heart Rate Variability and Heart Rate Recovery. Med Princ Pract. 25: 374–377, 2016.

[4] Gaur P, Bose S, Evaluation of Autonomic Responses at Different Phases of Menstrual Cycle with Reference to Body Mass Index. National Journal of Basic Medical Sciences. 9(1): 35-43, 2018.

[5] Padmanabha P, Rahmath A, Kote N, Goothy SSK & Deepa PK, Correlation of pupil to limbus diameter ratio (PLD ratio) with blood pressure and pulse rate in underweight females. International Journal of Health Sciences. 6(2), 11644 -11648, 2022.

[6] De K, Chatterjee S, Patra S, Mishra J, A study of sympathetic autonomic function in different phases of menstrual cycle among young adult females. Natl J Physiol Pharm Pharmacol. 9(7): 579-582, 2019.

[7] Mojumder DK, Patel S, Nugent K, Detoledo J, Kim J, Dar N et al, Pupil

to limbus ratio: Introducing a simple objective measure using two-box method for measuring early anisocoria and progress of pupillary change in the ICU. J Neurosci Rural Pract. 6(2): 208-215, 2015.

[8] Goothy SK, Chouhan RS, Garg N, Choudhary A, Mahadik KV, Correlation of pupil to limbus diameter ratio with blood pressure and heart rate among Parkinson's disease patients. Journal of Clinical and Diagnostic Research. 16(7): 16-19, 2022.

[9] Sagadevan YS, Parapurath S, Goothy SK, Choudhary A, Chouhan RS, Association of pupil-to-limbus diameter ratio with blood pressure and pulse rate in type 2 diabetic patients. Asian Journal of Medical Sciences.13(12): 123-126, 2022.

[10] Bhagyashree N, Ramaswamy C, Abeetha S, Sureka V, Ganesh M, Suma S, Comparison of anthropometric and physiological hemodynamic parameters in metabolic syndrome population. Natl J Physiol Pharm Pharmacol. 12(03): 277-280, 2022.

[11] Archana R, Sailesh KS, Bassetti S, Mishra S, The relationship between blood pressure and pupil to limbus diameter ratio in hypertensive women: a pilot study. Asian J Pharm Clin Res. 10(11): 142-144, 2017.

[12] Bindiya RS, Kishan K, Damodara GKM, Ramaswamy C, Ramesh Bhat M, Association between the different phases of menstrual cycle and time domain analysis of heart rate variability in young adult and older adult in Indian women. RJPBCS. 2(1): 297-303, 2011.

[13] Tkachenko NM, Il'ina EM. Akush Ginekol, Autonomic nervous system activity during the normal menstrual cycle. Akush Ginekol. (1): 35-38, 1994.

[14] Mendelsohn ME, Karas RH, Science. 308: 1583 -1587, 2005.

[15] Oyster CW, The Iris and the pupil. The Human Eye: Structure and Function. Sunderland, Mass: Sinauer Associates. 411-446, 1999.

[16] Walker HK, Hall WD, Hurst JW, Clinical Methods: The History, Physical, and Laboratory Examinations. 3rd ed. Ch. 58. Boston, MA: Butterworths. 1990.

[17] Bradley MM, Miccoli L, Escrig MA, Lang PJ, The pupil as a measure of emotional arousal and autonomic activation. Psychophysiology. 45(4): 602-607, 2008.