Original Article

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Physiology Section

Choice Reaction Time in Different Phases of the Menstrual Cycle in Healthy Females: A Cross-sectional Study

PARIKSHIT MULEY[1], PRANJALI MULEY[2], PURVA LANJEKAR[3], PUJA LANJEKAR[4], SHRUTIKA TATHOD[5], SURENDRA WADIKAR[6]

ABSTRACT

Introduction: The gonadal hormone output of females fluctuates monthly, and the endometrium, ovaries, and other genital organs undergo physical changes. It is known that women experience fluctuations in mood, energy, and cognitive abilities during different phases of their Menstrual Cycle (MC). Reaction Time (RT) is utilised to measure the ability of brain processing. The present study analysed the effect of various phases occurring in the MC on choice RT.

Aim: To determine whether hormonal changes that occur throughout the MC and its phases may have any effect on cognition.

Materials and Methods: An observational analytical cross-sectional study was conducted in the Department of Physiology at Jawaharlal Nehru Medical College, Wardha, Maharashtra, India. The study duration was six months, from August 2021 to April 2022. A total of 50 apparently healthy females aged 18-25 years were included in the study. The subjects with a history of normal and regular MC for the last six months were included. The Choice Reaction Time (CRT) with visual and auditory stimuli was analysed using an RT apparatus developed by Anand Agencies, Pune, India. In CRT, random auditory and visual stimuli were given to the subjects, and they had to react accordingly. The RT apparatus timer started when the stimuli were given, and it automatically stopped when the subject reacted, providing the RT. RT was noted in the subjects on four occasions: two days prior to menstrual bleeding (premenstrual), the 2nd day during menstruation, the 8th day (follicular phase), and the 18th day after menstrual bleeding (luteal phase). The average value of each type of Visual Reaction Time (VRT) and Auditory Reaction Time (ART) in the various menstrual phases was noted and analysed. The level of significance in the various menstrual phases was analysed with inferential statistical study using one-way Analysis of Variance (ANOVA) and multiple comparison Tukey's post-hoc test.

Results: In the present study, prolongation of visual and ART was seen in the luteal phase. There was prolongation of VRT when the luteal phase was compared to the menstrual and follicular phase, but the difference was not statistically significant ($p<0.05$). While in ART, there was prolongation when the luteal phase was compared with the premenstrual, menstrual, and follicular phase, but the difference was not statistically significant ($p<0.05$).

Conclusion: The current study reveals that there was a non significant prolongation of visual and ART in the luteal phase of MC when CRT was analysed. Thus, it was observed that there was no influence of fluctuating levels of ovarian hormones on ART and VRT in various phases of the MC while conducting CRT.

Keywords: Auditory reaction time, Menstrual phases, Visual reaction time

INTRODUCTION

The MC is a complex process that involves several hormonal changes and physiological events. The monthly rhythmic variations in the sex hormones secreted during a female's reproductive years usually result in physical changes to the endometrium, ovaries, and other genital organs. This monthly rhythmic pattern is known as the female sexual cycle, or more commonly, the MC [1]. This activity is produced with the help of the hypothalamo-pituitary-ovarian axis [2].

The MC is divided into four phases: the follicular phase, ovulation, the luteal phase, and menstruation. The follicular phase begins on the first day of menstruation and lasts for approximately 14 days. During this phase, the Follicle Stimulating Hormone (FSH) stimulates the growth of follicles in the ovaries. Ovulation occurs around day 14 of the MC when the mature follicle ruptures and releases an egg. The luteal phase begins after ovulation and lasts for approximately 14 days. During this phase, the ruptured follicle forms the corpus luteum, which produces progesterone. If fertilisation does not occur, the corpus luteum degenerates, and menstruation occurs [3].

It is well known that women experience fluctuations in mood, energy, and cognitive abilities during different phases of their MC. Cognitive ability is measured by RT [1]. RT is an indicator of sensory-motor performance. It was employed in the present study as a technique to evaluate sensory-motor function [4]. RT is the amount of time between the stimulus and the subject's voluntary response [5]. It is useful to quantify the efficiency with which the central nervous system processes information and responds to it. Several studies have investigated the effect of the MC on RT in healthy females. Some studies have found that RT is faster during the follicular phase compared to the luteal phase [1,2,6]. Other studies have observed a prolongation of RT in the premenstrual phase [7,8]. Additional studies have found no significant difference in RT between the follicular and luteal phases [9,10].

A person's capacity to co-ordinate timely and appropriate responses to stimuli is measured by the Simple Reaction Time (SRT) test, which is typically regarded as a psychomotor test [4]. However, in the CRT task, random stimuli are presented, and the subject is required to respond accordingly. Thus, the subject needs to perceive the stimulus, identify, analyse, and decide on the proper motor response [4]. The stimuli in CRT are more complex and demand decisions about how to respond; thus, the test becomes much more cognitive [11]. In CRT tasks, a variety of stimuli were provided at random, and the individual had to react in accordance with the stimulus. CRT is a challenging procedure that entails selecting the best response by recognising, differentiating, and analysing the input. The novelty of the study is that it analysed the effects of

various hormonal changes occurring during menstrual phases on the cognitive performance of the central nervous system with the help of CRT.

MATERIALS AND METHODS

An observational analytical cross-sectional study was conducted in the Department of Physiology at Jawaharlal Nehru Medical College, Wardha, Maharashtra, India. The study duration was six months, from August 2021 to April 2022. A total of 50 apparently healthy female volunteers aged 18-25 years with a history of regular MC for the last six months were included in the study. The CRT with visual and auditory stimuli was analysed using an RT apparatus developed by Anand Agencies, Pune. Informed consent was obtained from the subjects who participated in the study. Ethical approval for the study was obtained from the Institutional Ethical Committee with reference number IEC/2020-21/8934 dated 08/07/2020.

Inclusion criteria: Subjects who provided consent and had regular menstrual cycles for the last six months were included in the study.

Exclusion criteria: Subjects with menstrual irregularities, dysmenorrhoea, use of any form of contraception (pills or devices), use of psychotropic drugs (sedatives, hypnotics, and tranquilizers), antihistamines, anti-epileptics, sleep disorders, any history of visual or hearing impairment, and any addictions such as smoking or alcohol consumption were excluded from the study.

Sample size calculation: The sample size was calculated using the following formula:

$$\text{Unlimited population: } n=\frac{z^2\times\hat{p}(1-\hat{p})}{\varepsilon^2}$$

$$\text{Finite population: } n'=\frac{n}{1+\frac{z^2\times\hat{p}(1-\hat{p})}{\varepsilon^2 N}}$$

Where:

z-z score (90% confidence level) -1.64

p-population proportion -50% (0.5)

e-margin of error - 10% (0.1)

N-180 (finite population size) (number of female medical students on campus)

$$n=\frac{(1.64^2\times0.5\times(1-0.5))}{(0.1^2)}=67$$

$$n'=67/\frac{(1+(67-1)}{180}\approx49$$ (sample size for the study)

Study Procedure

Visual Reaction Time (VRT): Red and green lights were used to provide the visual stimulus. Both the subject and the examiner were seated on opposite sides of an opaque partition. When the researcher pressed the switch, a fixed fore period of one second of visual stimuli was displayed on the front screen. When the individual responded to the stimuli by pushing a key, the RT apparatus stopped measuring time after a fore interval of one second. This time was recorded as VRT. Similarly, for the auditory stimulus (beep tone), the same procedure was followed to measure ART. For both the visual and auditory reaction speeds, the test was performed three times. The three VRT and ART readings were noted, and the lowest reading was considered for the analysis of the study.

In the above description, VRT and ART were mentioned as components of CRT, where the researcher randomly presented either a visual (red or green light) or auditory stimulus to the subject after a fixed fore period of one second. The subject had to respond by pressing the switch on their side. This task of randomly presenting a stimulus and reacting to it is referred to as CRT. The time interval between stimulus application and the response is measured as RT in milliseconds. The CRT was conducted on four occasions of the menstrual cycle: two days prior to menstrual bleeding (premenstrual), the 2nd day during menstruation, the 8th day (follicular/proliferative phase), and the 18th day after menstrual bleeding (luteal/secretory phase) [7].

STATISTICAL ANALYSIS

The average value of each type of CRT in different phases of the menstrual cycle (premenstrual, menstrual, follicular, and luteal) was noted and analysed. Statistical analysis was performed using descriptive and inferential statistics, including one-way ANOVA and the multiple comparison Tukey's post hoc test. The software used for the analysis was Statistical Package for Social Sciences (SPSS) version 27.0, and a significance level of $p<0.05$ was considered.

RESULTS

In [Table/Fig-1], the mean value of CRT by visual stimuli with the standard deviation in various phases of the MC is provided. When the one-way ANOVA test was applied to the descriptive data (as shown in [Table/Fig-2]), a significant difference in CRT to visual stimuli was observed. Furthermore, the data was statistically compared using the multiple comparison Tukey's post-hoc test (presented in [Table/Fig-3]), which revealed a statistically significant prolongation of VRT in the luteal/secretory phase compared to the premenstrual phase of MC. It was also noted that there was a prolongation of VRT when comparing the luteal phase to the menstrual and follicular phases, although the difference was not statistically significant.

Phase of MC	N	Mean	Std. Deviation	Std. Error	95% Confidence interval for mean	
					Lower bound	Upper bound
A	50	308.96	69.06	8.77	291.42	326.50
B	50	340.70	92.10	11.69	317.31	364.09
C	50	340.93	101.93	12.94	315.04	366.82
D	50	373.16	92.90	11.79	349.56	396.75

[Table/Fig-1]: CRT, VRT.
Comparison of Mean choice VRT score in four phases of MC
A: Premenstrual phase; B: Menstural phase; C: Proliferative/follicular phase; D: Luteal/secretory phase

Source of variation	Sum of squares	df	Mean square	F-value	p-value
Between groups	127750.37	3	42583.45	5.27	0.002, S
Within groups	1968882.83	244	8069.19		
Total	2096633.21	247			

[Table/Fig-2]: One-way ANOVA test showing comparison of mean choice VRT score in four phases of MC The one-way ANOVA tests showing- $p<0.05$ as significant (s).

Phase of MC		Mean difference (I-J)	p-value	95% Confidence interval	
				Lower bound	Upper bound
A	B	-31.74	0.203, NS	-73.47	9.99
	C	-31.96	0.198, NS	-73.70	9.76
	D	-64.19	0.001, S	-105.92	-22.45
B	C	-0.22	1.000, NS	-41.96	41.50
	D	-32.45	0.187, NS	-74.18	9.28
C	D	-32.22	0.192, NS	-73.96	9.50

[Table/Fig-3]: Multiple Comparison: Tukey's post-hoc test showing comparison of mean choice VRT score in four phases of MC.
Multiple Comparison: Tukey's test showing the comparison of VRT in four menstrual phases
A: Premenstrual phase; B: Menstural phase; C: Proliferative/follicular phase; D: Luteal/secretory phase; S: Significant; NS: Not significant

Similarly, in [Table/Fig-4], the mean value of CRT by auditory stimuli, along with the standard deviation in various phases of MC, is given. The one-way ANOVA test (depicted in [Table/Fig-5]) indicated no significant difference in ART among the phases of MC. Additionally, the multiple comparison Tukey's test (shown in [Table/Fig-6]) demonstrated

a prolongation of ART when comparing the luteal phase with the premenstrual, menstrual, and follicular phases, but the difference was not statistically significant.

Phase of MC	N	Mean	Std. Deviation	Std. Error	95% confidence interval for mean	
					Lower bound	Upper bound
A	50	309.59	83.94	10.66	288.27	330.91
B	50	318.87	98.70	12.53	293.80	343.93
C	50	317.72	107.88	13.70	290.32	345.12
D	50	330.72	142.67	18.11	294.49	366.95

[Table/Fig-4]: Comparison of mean Choice ART score in four phases of MC.
A: Premenstrual phase; B: Menstural phase; C: Proliferative/follicular phase; D: Luteal/secretory phase

Source of variation	Sum of squares	df	Mean square	F-value	p-value
Between groups	14095.33	3	4698.44	0.385	0.764, NS
Within groups	2975728.56	244	12195.60		
Total	2989823.89	247			

[Table/Fig-5]: One-way ANOVA test showing comparison of mean choice ART score in four phases of MC. The one-way ANOVA tests showing- p>0.05 as not-significant.

Phase of MC		Mean difference (I-J)	p-value	95% confidence interval	
				Lower bound	Upper bound
A	B	-9.27	0.966, NS	-60.58	42.03
	C	-8.12	0.977, NS	-59.43	43.17
	D	-21.12	0.711, NS	-72.43	30.17
B	C	1.14	1.000, NS	-50.16	52.45
	D	-11.854	0.933, NS	-63.16	39.45
C	D	-13.00	0.914, NS	-64.30	38.30

[Table/Fig-6]: Multiple Comparison: Tukey Test showing comparison of mean choice ART score in four phases of MC Multiple Comparison: Tukey test showing the comparison of VRT in four menstrual phases.
A: Premenstrual phase; B: Mensturalphase; C: Proliferative/follicular phase; D: Luteal/secretory phase; NS: Not significant

DISCUSSION

The objective of the current study was to observe whether sex hormones secreted across the menstrual phases affect cognitive functions, specifically RT. In present study, both visual and auditory stimulus-based CRT tests were administered to subjects in different phases of the MC.

In the CRT study, it was found that VRT was significantly prolonged in the luteal/secretory phase compared to the premenstrual phase of MC. However, when comparing the luteal phase to the menstrual and follicular phases, there was a prolongation of VRT, but the difference was not statistically significant. For ART, there was a prolongation in the luteal phase compared to the premenstrual, menstrual, and follicular phases, but again, the difference was not statistically significant.

These results are in agreement with a study conducted by Veena CN et al., which observed non significant differences in ART and VRT between the mid-secretory phase and proliferative phase [9]. Bhutkar MV and Aparna L noticed non significant differences in ART and VRT among the premenstrual phase, post-menstrual phase, and menstrual phase of MC [10]. Donadi A et al., also observed no statistically significant influence of oestrogen and progesterone on reaction time (ART and VRT) between the premenstrual and post-menstrual phases of MC [11].

However, some other studies have shown a prolongation of reaction time in the luteal phase. Kumar S et al., observed delayed ART and VRT in the luteal phase compared to other phases [1]. Patel J et al., concluded that ART is increased in the luteal phase [6]. Garg R et al., observed a significant delay in reaction time during the luteal phase compared to other MC phases [2].

On the other hand, some studies have observed a prolongation of reaction time in the premenstrual phase. Nene AS and Pazare PA noticed a significant increase in ART during the premenstrual phase [7]. Sudheer C et al., observed a significant prolongation of both ART and VRT during the menstrual phase [8]. Patel J et al., observed a significant prolongation of both simple VRT and choice VRT in the premenstrual phase [12].

The main cause of increased RT in the luteal phase is the influence of sex hormones, primarily progesterone, which leads to the retention of salt and water. This hormonal influence affects axonal conduction, causing a prolongation of RT [2]. The delayed axonal conduction reduces impulse conduction and increases RT by altering the availability of neurotransmitters at synapses [13]. Oestrogen decreases the synthesis of Gamma-Aminobutyric Acid (GABA), leading to increased activation of glutamate receptors. Progesterone decreases neuronal excitability and improves GABA neurotransmission. The varying levels of oestrogen and progesterone throughout the premenstrual and postmenstrual phases contribute to the more prolonged RTs in the premenstrual phase [11,14]. Additionally, the menstrual cycle can cause changes in corneal thickness, with the thinnest point at the cycle's beginning and the thickest at the end. This might be a reason for diminished eyesight and prolonged VRT during the premenstrual phase [11,15].

The strength of the present study lies in the use of CRT tests instead of SRT tests employed in other similar studies. SRT tests are typically considered psychomotor tests, while CRT tests involve more complex stimuli that require analysis and decision-making in response [4,16]. Consequently, CRT tests are more cognitively demanding [4,16]. Therefore, CRT is more sensitive to latency as additional time is required for cognitive stimulus analysis [16,17]. The present study revealed a non significant prolongation of visual and auditory reaction times during the luteal phase when analysed using CRT. The observations suggest that fluctuating levels of ovarian hormones do not significantly influence CRT with auditory and visual stimuli during various phases of the menstrual cycle. The study's findings indicate that cognitive performance is not greatly influenced by different phases of the menstrual cycle. In future research, including other cognitive function tests related to attention, perception, memory, intelligence, and language could provide further insights into cognitive functions.

Limitation(s)

The study had several limitations. Firstly, it was a cross-sectional study with a restricted age group of 18-25 years, which limited the generalisability of the study's conclusions to the entire population. Additionally, increasing the sample size would help reduce errors in the study group and enhance the reliability of the findings. Furthermore, the study focused solely on the CRT test as the parameter for assessing cognitive function. Future research could consider incorporating other cognitive testing parameters to investigate the effects of hormonal changes on cognition.

CONCLUSION(S)

There was a statistically significant prolongation of VRT in the luteal/secretory phase compared to the premenstrual phase of the MC. However, when comparing the luteal phase to the menstrual and follicular phases, there was a prolongation of VRT, but the difference was not statistically significant. During the CRT for auditory stimulus, there was a prolongation of ART in the luteal phase compared to the premenstrual, menstrual, and follicular phases, but again, the difference was not statistically significant. The study suggested that there is a non significant prolongation of CRT in the luteal phase of the MC, indicating that fluctuating levels of ovarian hormones do not significantly influence CRT with auditory and visual stimuli during various phases of the MC.

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PARTICULARS OF CONTRIBUTORS:
1. Professor, Department of Physiology, Datta Meghe Medical College, Nagpur, Maharashtra, India.
2. Associate Professor, Department of Physiology, Datta Meghe Medical College, Nagpur, Maharashtra, India.
3. Undergraduate Student, Department of Physiology, Jawaharlal Nehru Medical College, Wardha, Maharashtra, India.
4. Undergraduate Student, Department of Physiology, Jawaharlal Nehru Medical College, Wardha, Maharashtra, India.
5. Undergraduate Student, Department of Physiology, Jawaharlal Nehru Medical College, Wardha, Maharashtra, India.
6. Additional Professor, Department of Physiology, BYL Nair Charitable Hospital and Topiwala National Medical College, Mumbai, Maharashtra, India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:
Dr. Pranjali Muley,
N-20, Sneh Nagar, Wardha Road, Nagpur-440015, Maharashtra, India.
E-mail: drpranjali11@gmail.com

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