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Evaluation of the parasympathetic neural outflow to the heart during different phases of menstrual cycle as assessed by heart rate variability: a cross-sectional study

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Abstract

Decrease in parasympathetic neural activity has been a predictor for development of diseases like cardiac arrhythmias, hypertension etc., measuring the vagal outflow in humans using invasive techniques is difficult which has paved way for Heart rate variability (HRV) which is anoninvasive tool for assessing periodic and non-periodic variations in HR *which in women is affected by endogenoussex hormones. The aim was to compare the cardiac parasympathetic tone during follicular and luteal phases of menstrual cycle using the frequency and time domain parameters of HRV. Thirty female students aged 17-20 years were subjected to ECG recording which was fractionated analogally into Time and Frequency Domain Analysis during follicular and luteal phases of menstrual cycle. Data was analyzed statistically using student's pair t-test. The difference in Mean HR, Rmssd, NN5o, pNN50 and HF nu was found to be statistically significant in follicular and luteal phase. Estrogen during the follicular phase of menstrual cycle promotes vagal outflow while the Progesterone during the luteal phase promotes sympathetic activity hence we conclude that the sex hormones during menstrual cycle interact with ANS affecting the regulation of the heart.*

Keywords: HRV, Vagal activity, HF nu.

Introduction

Throughout the biological lifespan of a female there are rhythmic cyclical changes which are the physiological preparation by the female body for fertilization and conception[1]. The duration of menstrual cycle averages from around 28 to 30 days but may show variability[2]. The menstrual cycle in females is characterized by the follicular and luteal phases which represent the cyclical changes occurring in the ovaries. The ovarian follicles develop following the bleeding phase during which levels of hormone estrogen gradually increase while progesterone levels remains low and this is known as Follicular phase. But the ratio is reversed in favor of the hormone progesterone in luteal phase as compared to estrogen[3].

The two divisions of the peripheral nervous system i.e.- sympathetic and parasympathetic jointly modulate the working of the heart on beat-to beat basis and its rhythm which is controlled by a pacemaker the SA node[1,2]. Under normal resting conditions the SA node receives tonic discharges from both the sympathetic as well as parasympathetic nervous system but there is suggested parasympathetic dominance since the effects of vagal stimulation decay quickly and has a brief latency which suggests the role of parasympathetic activity primarily in the regulation of SA node discharge. Heart Rate Variability is anon-invasive technique to measure variations in RR intervals between consecutive beats[4-6]. Availability of computer interface has made it easier to study the beat-to-beat variations of the sinus nodal discharge and power spectral analysis of HRV[7].

The two algorhytmes of measurement of HRV include the Fast Fourier technique and Auto regression[4,5]. LF and HF power which represent the FFT measure the behaviors of the two arms of the ANS. The HF component measures the vagal modulation on the heart which shows a frequency discharge in the range (0.15-0.4 Hz) whereas the LF component points towards the interaction of combined action of sympathetic as well as parasympathetic neural activity but more so of the sympathetic activity and shows frequency discharge of (0.04-0.15); while LF / HF ratio is a global indicator of sympathovagal activity. A reduced HRV denotes autonomic imbalance either in the form of decreased parasympathetic activity or increase in the activity of the sympathetic activity and is considered as important risk factor for prediction of cardiac catastrohies[4,5]. Time domain measures used in this study are SDNN, pNN50 and Rmssd. The SDNN component reflects the combined activities of sympathetic and parasympathetic systems while pNN50 and Rmssd reflect the influence of parasympathetic innervation on the pacemaker. Reduced SDNN is considered a reflection of sympathovagal imbalance[4,5].

Studies conducted on animal experimentation have proved that estrogens act centrally to influence the vagal outflow increasing it and decreasing sympathetic activity[8], thus providing a cardiovascular protective function. Progesterone, on the other hand, appears to have an opposing effect both at the central as well as peripheral levels, elevating central noradrenalin release[9,10]. Given this antagonism, hormonal fluctuations across the cycle is associated with changes in peripheral nervous system functioning. Low parasympathetic activity is inculcated in development of future disease[11,12]. There are several studies suggesting a definitive role of HRV changes across the menstrual cycle[13]. There are very few studies to cite which have focused on the variations in the neurocardiac parameters during the various phases of menstrual and the extent to which these endogenous gonadal hormones actually interact with the cardiovascular functions and are a matter of interest as well as conflict due to multiple contradictory reports of the same. Hence it is mandatory for identification of HRV variations in women at risk to develop heart disease so as to permit an early intervention.

Materials and methods

Study Design: Cross-sectional.

Duration: December 2011 till February 2012.

Source of data: In the present study the data was collected from the students of first year M.B.B.S enrolled in J. N. Medical College, Belgaum for the academic year 2011.

Sample size: Thirty eumenorrhic girls aged 17-20 years of age studying their MBBS course at J.N.M.C, Belgaum were selected

Sample size estimation -
Expected Reduction-(Mean) =d=20
SD=40 = σ
$ά = 0.05$ One sided $Z ά = 1.65$
$β = 0.2$ Power 80% $Z β = 0.84$
$n = [(Z ά + Z β) σ / d]^2 = 24.8 = 25 = 30$

Based on sample size calculation 30 MBBS first phase females aged 17-20yrs of J.N.M.C who are eligible were enrolled at the time of data collection.

Inclusion criteria: Thirty eumenorrhic girls aged 17 -20 years of age studying their MBBS course at J.N.M.C, Belgaum with regular menstrual cycle duration of 28-30 days duration at least 3 months prior to the study, having no medical or gynecological problems, no well-defined premenstrual tension were enrolled.

Exclusion criteria: Subjects who had irregular menstrual cycles, any endocrine disorders, consuming medications likely to affect or influence PMS (diuretics, etc.) or drugs affecting moods (antidepressant, tranquilizers), oral contraceptive pill, hormonal replacement therapy, drugs that alter the cardiovascular functions were also excluded from the study.

Voluntary Informed written consent was obtained from all participants, and the experiment protocol was approved by Institutional Ethics committee of the college. In every case selected, thorough menstrual history was taken including nature and days of menstrual flow, regularity and total duration of cycle as well as detailed medical history was obtained from all participants, and they underwent a standard physiological examination.

The entire purpose of the study and a detailed description of the study protocol given to all the subjects prior to beginning of the battery of tests and examination carried out at same time of the day to avoid diurnal variations. The battery of tests used to study the parasympathetic responses during different phases of menstrual cycle were carried out during follicular and luteal phases of menstrual cycle. Considering the first day of bleeding as Day 1 and basal body temperature, the phases were marked out. All female subjects were studied at each of the following phases during a single menstrual cycle: i. Follicular (10^{th} day), ii. Luteal (20^{th} day).

Study procedure: i. The subjects were asked to report to the lab in the morning at around 8.00 am, avoided caffeinated drinks, alcohol or nicotine prior to the experiment and completed their evening meal by 9 P.M. Students avoided any sort of heavy physical activity on the day of the test or the day prior. ii. ECG Recording: The placement of the ECG electrodes was using the conventional Lead wherein the electrodes were placed on RA (right arm), LA (left arm), LL (left leg), and RL (right leg). Heart rate Variability analysis was done using Naviquire Computerized ECG system (ECG V; 52 Manufactured by NIVIQURE Meditech pvt. Ltd. Bangalore and marketed by Inco Medicals; Ambala (Manufactured year-2006). The ECG signal was first analogally recorded and then digitally converted and analyzed in the frequency domain and time domain parameters. The analog ECG signal was obtained using lead II to obtain a QRS complex of sufficient amplitude and stable base line. Occasionally, on every record, noise (appearing as pairs of short intervals due to a false trigger that resulted from the movement of the subjects) was removed, and a small number of too-long R-R intervals at the onset of the record were also removed. Finally, the remaining R-R intervals, containing were stored on a computer for analysis offline. iii. The HRV is recorded for duration of 5 minutes with the subject lying supine. In the frequency domain analysis the HF component relates to the activity in the parasympathetic arm while the same is represented by SDNN, Rmssd, NN50, pNN50 components in the Time Domain measurement[13]. HF values are represented in normalized unit (nu) or as absolute unit i.e.-ms.

Analysis plan: The data was represented as mean ± SD using software SPSS 18. The statistical analysis was conducted using Student's paired T-test and a value of $p<0.05$ was considered significant

Results and discussion

In the current study we have measured the variability using HRV parameters in both the domains in Follicular and Luteal phases of healthy young females. It was evident from the results that HF (peak) and HF (nu) which are frequency domain measures of parasympathetic activity are higher during the follicular phase as compared to lutealphase, while Rmssd, NN50, PNN50 which relate to the same in the time domain measure are lower in the luteal phase. The reduced values of SDNN in the second half of the cycle i.e. luteal phase points towards an imbalance between the two divisions of the autonomic system as this phase is characterized by increasing levels of the hormone progesterone in comparison to the follicular phase where estrogen tends to rise. From our overall results it is quite evident that the vagal activity is altered by increasing amounts of progesterone in the luteal phase of young females.

Table-1: Demographic details.

Age (years)	18.3±0.66 (17-20)
Weight (kg)	56.7±7.55 (47-85)
Height (m^2)	1.56±0.04 (1.51-1.68)
Body Mass Index (kg/m^2)	23.12±3.23 (18.59-35.38)

Table-1 demonstrates the demographic distribution of participants, with the mean age of the participants being around 18.3 years, weight (56.7±7.55), height (1.56±0.04) and BMI (23.12±3.23).

Table-2: Frequency Domain parameters of HRV analysis during follicular and luteal phases.

	Follicular (Mean±SD)	Luteal (Mean±SD)	P value
HF(peak)(ms)	2846 ±186	1743 ±113	0.001
HF(nu)	54.1 ±1.07	49.6 ±1.48	0.001
LF/HF Ratio	0.8 ± 0.044	1.05 ±0.043	0.001
HR(bpm)	78.8 ±7.48	84 ± 8.17	0.003

Table-2 shows that the HF (peak) as well as HF (nu) both representing the absolute and normalized values of the parasympathetic neural activity was statistically higher in the follicular phase. Luteal phase was characterized by greater LF/HF ratio which indicates reduced parasympathetic activity, increased sympathetic activity or a combination of both. The resting heart rate was statistically higher during the luteal phase proving a sympathetic dominance during this phase.

Figure-1 Showsa higher HF (nu) during the Follicular phase which is a marker of parasympathetic neural activity.

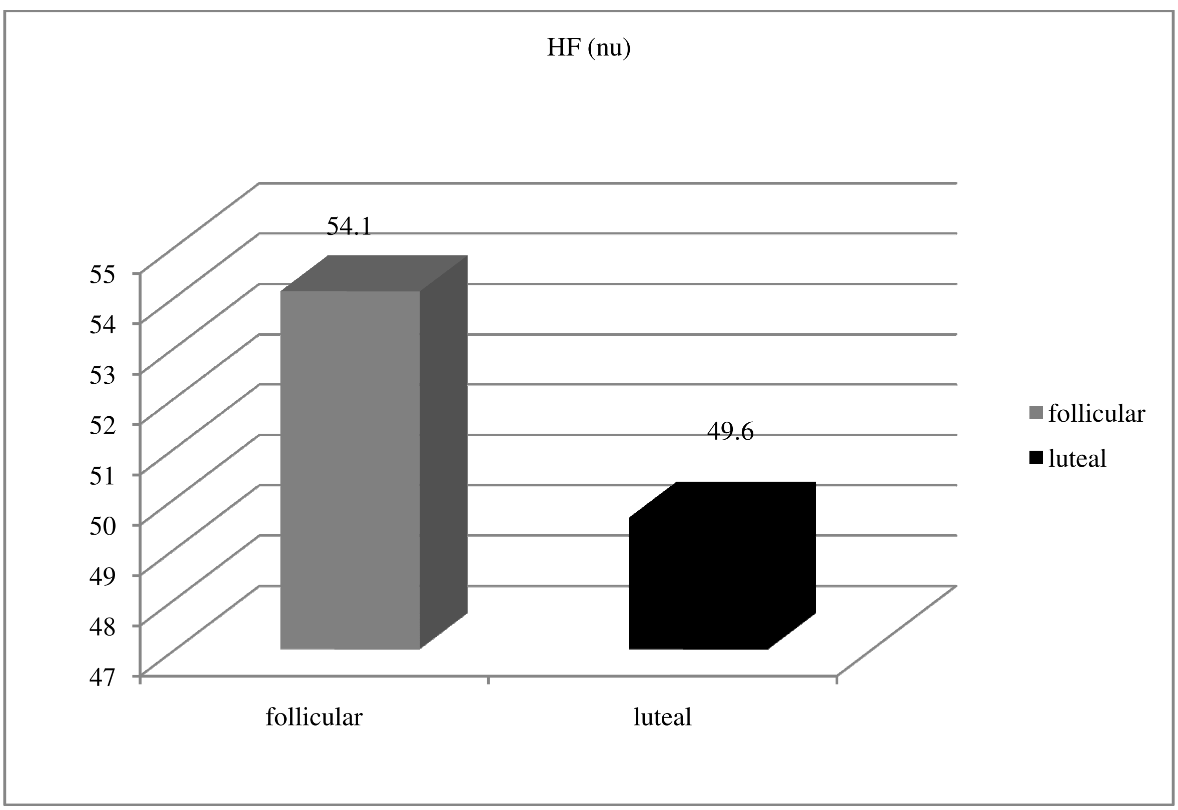

Figure-1: HF (nu) during follicular and luteal phases.

Figure-2 shows greater sympathetic tone across the luteal phase resulting in higher resting heart rate.

Table-3: Time Domain parameters of HRV analysis during follicular and luteal phases.

	Follicular (Mean±SD)	Luteal (Mean±SD)	P value
SDNN (ms)	164 ±32	136±39	0.001
Rmssd (ms)	30.8 ±1.92	27.23 ±1.62	0.001
NN_{50} (ms)	12.30 ±7.69	7.62 ±6.69	0.001
pNN_{50} (%)	8.70±5.63	5.32±4.82	0.001

Table-3 shows the values of the time domain variables SDNN which reflects a combined activity of sympathetic and parasympathetic activity while Rmssd, NN50 and pNN50 which individually or together reflect the parasympathetic control on the SA node. Table-2 shows reduced values of SDNN as well as Rmssd, NN50 and PNN50 in the luteal phase.

Figure-3 SDNN which is a marker of sympathovegal activity is reduced in the luteal phase which points towards Sympathovagal imbalance.

Figure-4 shows the vagal activity markers in the time domain analysis i.e.-Rmssd, NN50, pNN50 which are all reduced in the luteal phase denoting a reduced activity during the same.

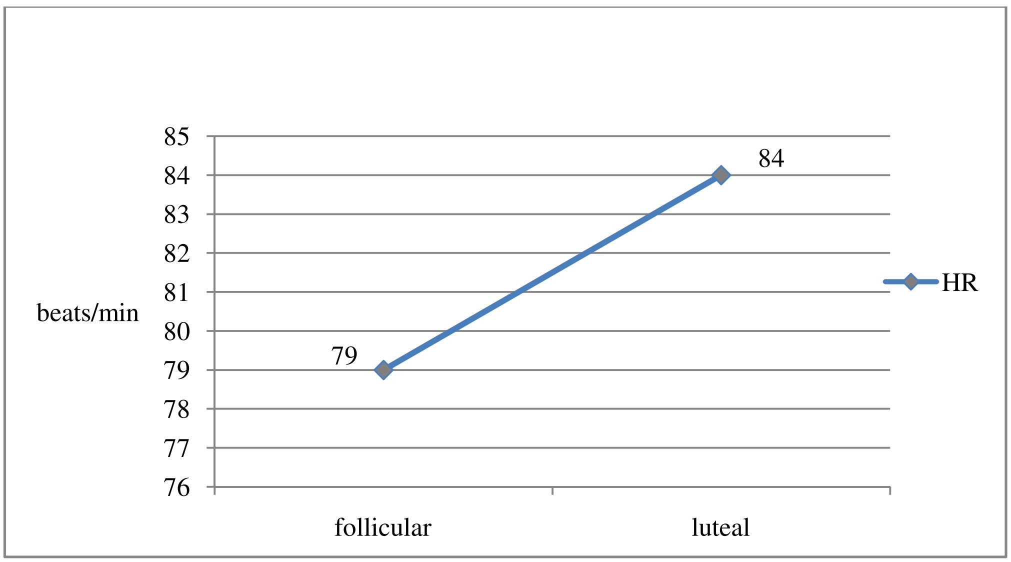

Figure-2: Resting HR across Follicular and Luteal phase.

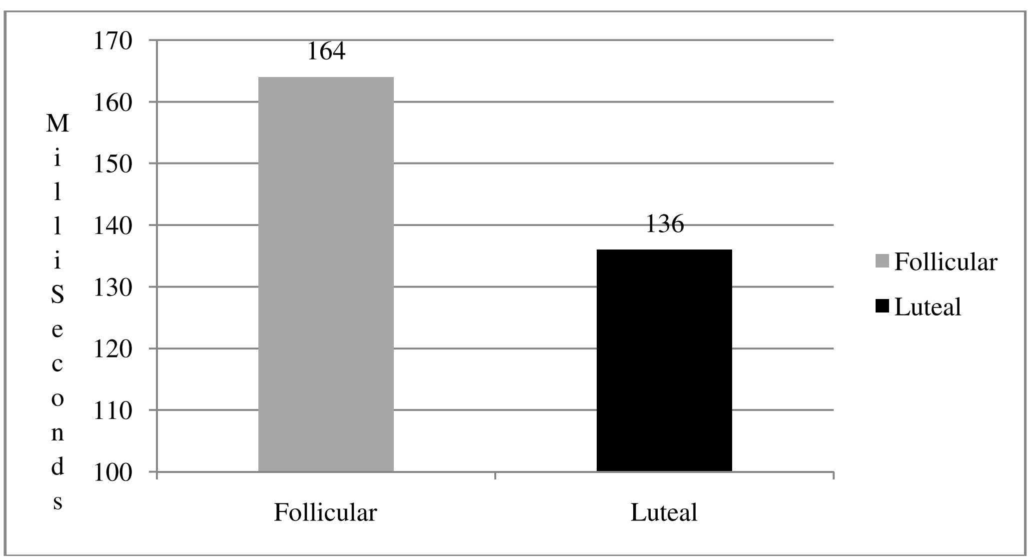

Figure-3: SDNN across the phases.

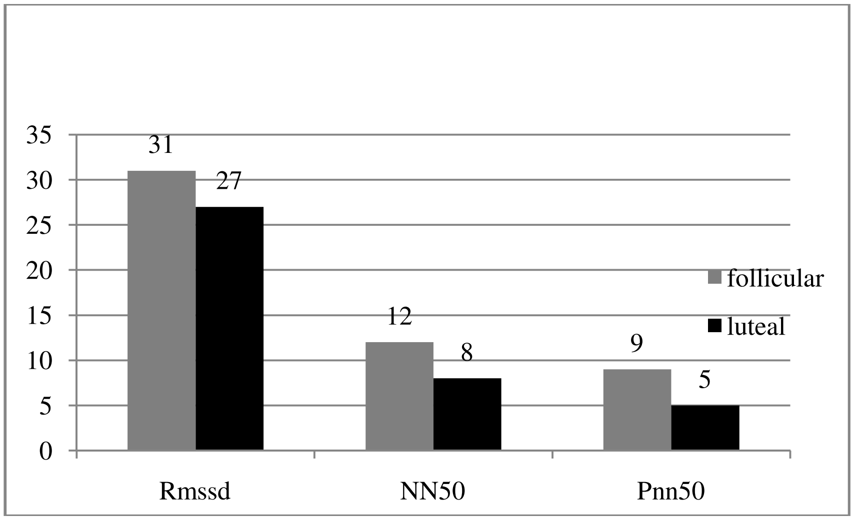

Figure-4: Time domain variables denoting the parasympathetic neural activity.

Discussion: In 1983 a study conducted by Goldstein et al suggested the possible interaction of endogenous sex hormones of menstrual cycle and the ANS[14]. The first clinical study supporting the above was reported by Sato et al who investigated 20 college students during the follicular phase and luteal phase and their results showed an increased LF nu, decreased HF nu and increased LF/HF ratio in luteal phase indicating predominant sympathetic activity during luteal phase[15].

In the present study a comparison was made with regards to the resting heart rate and heart rate variability especially parasympathetic parameters both the time and frequency domain variables during follicular and luteal phases of menstrual cycle. The present study showed a higher HF, Rmssd, NN50, pNN50 (Vagal indices) during the follicular phase which could possibly be due to increase in the levels of cardio-protective estrogen, whereas higher resting HR, frequency domain variables like LF, LF/HF ratio (sympathovegal imbalance) as well as reduced SDNN suggest a sympathetic predominance during the luteal phase which may be the result of high progesterone levels during the same.

Series of studies have demonstrated that the luteal phase is associated with a significantly reduced parasympathetic activity which is measured by the HF component, reflecting itself as altered LF/HF ratio. The high progesterone levels during the luteal phase may have an inhibitory role on cardio-vagal activity[14-16]. A study conducted has demonstrated changes in the absolute values of HRV but the results seem to be unequivocal[17]. The results were congruent with a study done where they showed a higher sympathetic activity and a sympathovagal imbalance in luteal phase with an associated increase in parasympathetic activity in the follicular phase but the results were not equivocal[18]. Series of studies conducted have concluded the luteal phase as to showing sympathetic predominance[19-22], studies done at molecular levels have indicated that centrally actions of estrogens include facilitation of cholinergic transmission and activating the synthesis of a vital rate limiting enzyme choline acetyltransferase, involved in the formation of acetylcholine formation , and also have a role in sympathetic outflow inhibititon[23,24]. In physiological doses progesterone acts as a sympathomimetic agent which induces nor epinephrine release[25,26]. Controversies still exist, a study conducted has reported diametrically opposite findings with increased high frequency (HF) values during the luteal phase[27]. Due to inconsistencies in the results reported by various studies and little availability of the information regarding the interactions of sex steroids and HRV in literature this study was conducted.

Conclusion

Hence we concluded that parasympathetic neural activity as a component of autonomic nervous system is affected by different phases of menstrual cycle & it deems likely that endogenous female hormones exert their effects on the peripheral neural system which is reflected by the cardiovascular system functioning. This finding has clinical implications since the luteal phase which shows a sympathetic predominance and sympathovagal imbalance should be viewed and interpreted with concern as a phase for predilection for cardiac autonomic dysfunction.

Limitations and future proposal: Due to technical and methodological restraints actual hormone estimate and its effect on cardiac autonomic tone could not have been undertaken. Future studies should be extended to conditions like hypertension and other cardiovascular disorders to understate the alterations that underlie these diseased conditions.

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