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Determination of Physical Fitness Index in First Year Medical Students by Harvard Step Test

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ABSTRACT

Physical fitness is taken as one of the most important parameter in evaluating a person's ability to perform strenuous activities like sports, military forces, etc. The following study was done to determine the physical fitness among first year boys who were physically active and those who lead a sedentary life. Material- 60 male students taken from among the first year students, divided into two groups of 30 subjects each. One group contains only physically active students and the other group contains only sedentary students. They were made to exercise on the Harvard Step Test and their Physical Fitness Index was calculated. Result- It was seen that the group with physically active subjects showed better result in comparison to sedentary group. Conclusion- Harvard Step Test can be used as a good measure of cardiovascular fitness.

Keywords: *Physical Fitness, Physical Fitness Index, Physically Active, Sedentary, Harvard Step Test*

INTRODUCTION

In order to utilize human power in best possible and most economical way, it is essential to know the maximal physical ability level of an individual and the factors limiting it. Physical fitness can be defined as quantitative expression of physical condition of an individual. Physical fitness of an individual varies with age and is maximum at 20-30 years[1]. Now a days more emphasis was put on Physical fitness aspect in many fields like sports, training programme in military forces, patients suffering from cardio-respiratory diseases. For the purpose of quantitative assessment one's physical ability, various methods are there, out of which the Harvard fatigue step test[2] was performed.

MATERIAL AND METHOD

The present study was conducted in Department Of Physiology, S.C.B Medical College, Cuttack from period of May 2004 to September 2006 by taking 60 male subjects from the first year students within age group 17-21 years. The height of the students range from 160 to 174 cm and weight 43-72 kg.

The subject were divided into two groups, i.e. Physically Active Group which include students who regularly took active part in either atheletics or any other sports and Sedentary Group which include subjects who do not perform any physical work.

Exclusion criteria include subjects who were physically handicapped or having cardio-respiratory disorders. The subjects were demonstrated the procedure. First their pre-exercise Pulse Rate (PR), Systolic Blood Pressure (SBP) and Diastolic Blood Pressure (DBP) were recorded. Then each of them has to go up and down the Harvard Step, completing a cycle in 2 second for a period of 5 min or till he gets exhausted, whichever is first.

The duration of exercise was recorded by help of a stop-watch. After exercise he was made to sit comfortably on chair and his post-exercise PR, SBP and DBP were recorded. Recovery PR was recorded for 30 sec each in 1 to 1.5 min, 2 to 2.5 min and 3 to 3.5 min.

Physical Fitness Index was calculated by the formula[3]

$$PFI = \frac{\text{Total duration of exercise in seconds}}{2(\text{Sum of } \tfrac{1}{2} \text{ min PR at 1 min+2 min+3 min after exercise})}$$

Statistics used was Unpaired t-test. Software used was SPSS 16.0.

FINDINGS

The resting values of PR, SBP and DBP were first recorded in the 60 subjects. They were then instructed to perform the Harvard Step Test for 5 min or toll exhaustion, whichever was first reached. Then the post exercise PR at 1, 2 and 3 minutes were recorded. The post exercise SBP and DBP at 1 minute were also recorded. The following values were observed:-

Table 1: Shows comparison of the age, height and weight between the two groups

PARAMETERS	GROUP	MEAN	STD. DEV	t	p
Age (in years)	Sedentary	20	1.11	0.38	0.70
	Physically active	19.88	1.09		
Height (in cm)	Sedentary	163.28	5.45	1.36	0.17
	Physically active	160.92	6.69		
Weight (in Kg)	Sedentary	53.9	5	1.614	0.113
	Physically active	50.84	4.41		

'p' calculated by Unpaired t- test.

Table 2 : Comparing the PR, SBP, DBP in both groups both before and after exercise

PARAMETERS	GROUP	MEAN	STD. DEV	t	p
Pre-exercise PR (beats/min)	Sedentary	72.84	2.26	3.06	0.003
	Physically active	70.96	2.07		
Pre-exercise SBP (mmHg)	Sedentary	119.36	4.99	0.31	0.75
	Physically active	119.76	4.01		
Pre-exercise DBP (mmHg)	Sedentary	81.76	2.60	2.87	0.005
	Physically active	78.8	4.43		
Post-exercise PR_{1min} (beats/min)	Sedentary	66.6	9.16	3.34	0.001
	Physically active	59.04	6.62		
Post-exercise PR_{2min} (beats/min)	Sedentary	60	7.72	4.37	6.51
	Physically active	51.88	5.14		
Post-exercise PR_{3min} (beats/min)	Sedentary	54.48	9.43	4.81	1.52
	Physically active	43.52	6.37		
Post-exercise SBP (mmHg)	Sedentary	148.96	14.35	2.77	0.007
	Physically active	138.68	11.65		
Post-exercise DBP(mmHg)	Sedentary	68.56	4.6	3.78	0.004
	Physically active	62.8	6.05		

'p' calculated by Unpaired t- test.

Table 3: Comparing the Physical Fitness Index (PFI) score and Endurance in both groups

PARAMETERS	GROUP	MEAN	STD. DEV	t	p
PFI score	Sedentary	50.25	10.04	10.59	<0.001
	Physically active	90.62	16.18		
Endurance(Sec)	Sedentary	179.4	31.96	10.44	6.01
	Physically active	275.2	32.89		

'p' calculated by Unpaired t- test.

CONCLUSION

The subjects in both the groups were comparable on the basis of age (p=0.7), height (p=0.17), and weight (p=0.113). The resting PR of sedentary group is significantly higher than physically active group (p= 0.003). The resting SBP of both the groups were comparable (p=0.75) but resting DBP of sedentary males were found to be significantly higher than that of physically active group (p=0.005).

Exercise done using Harvard Step Test with stepping heights of 18 inches and done for 5 min or till exhaustion, whichever is first. It had been proven effective by Gollanger and Brouhe[4] and Johnson et al.

The post exercise PR at 1min was seen to be significantly low in physically active male (59.04 beats/min) than sedentary males (66.6 beats/min). This is similar to the study of Bandopadhyaya[5].

The post exercise SBP and DBP were found to be significantly higher in Sedentary group (148.96mmHg & 68.56mmHg respectively) than in Physically active group(138.68mmHg & 62.8mmHg respectively). This findings coincided with the work of Bandopadhyaya[5] and Bernard[6].

The Physical Fitness Index was found to have indirect correlation with weight and Pulse Rate and direct correlation with body height. It corresponds to the work of Bandopadhyaya et al[7], Astrand[8] and Elbel[9] respectively. In this study the PFI Score is significantly higher in Physically Active group(50.25) than Sedentary group(90.62) but the Endurance shows insignificant high level in Physically Active group.

The Harvard Step Test is a good measure of cardiovascular fitness, the physical fitness index of physically active students is higher than sedentary students having approximately equal body parameters of same age group in both sexes.

ACKNOWLEDGEMENT

I want to thank my Associate Professors, Dr. Karuna Dash and Dr. Nirupama Ray and all my colleagues for their help during the entire period of my study.

Ethical Clearance: The study was conducted in the Department of Physiology after receiving approval from the Institutional Ethics Committee of S.C.B Medical College, Cuttack, Utkal University.

Source of Funding: Self

Conflict of Interest: Nil

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Effect of Alternate Nostril Breathing on Cardiac Output and Systemic Peripheral Resistance

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ABSTRACT

Alternate nostril breathing (ANB, Anulom-Vilom) is one of the most popular pranayama yogic exercises. The present study was done to find out direct effect of alternate nostril breathing on cardiac output and peripheral resistance by using Impedance Cardiovasograph (Nivomon, L&T Medical's). One hundred asymptomatic healthy male subjects, aged 17-23 years, participated voluntarily in the present study. Cardiac output, systemic peripheral resistance and other cardiovascular parameters were measured before and after Alternate Nostril Breathing (ANB) exercise of 15 minutes. Statistically significant decrement was observed in all cardiovascular parameters after ANB exercise but decrement was more pronounced in diastolic blood pressure (DBP), heart rate (HR) , Systemic Peripheral Resistance (SPR) and Systemic Vascular Resistance Index (SVRI) incomparison to decrement in systolic blood pressure (SBP), cardiac output (CO), stroke volume (SV), cardiac index (CI), stroke volume Index (SI).

Keywords: *Alternate Nostril Breathing, Impedance Cardiovasograph, Cardiac Output, Systemic Peripheral Resistance*

INTRODUCTION

Pranayama is a yogic discipline of ancient India. Pranayama includes different respiratory techniques. Anulom-Vilom, or alternate-nostril breathing (ANB) is a type of pranayama that involves alternation in nostril breathing. It may alter cardio respiratory and autonomic parameters. Several investigations have been conducted to determine the long-term effects of this technique on the cardiovascular and autonomic nervous systems in healthy and clinical populations and many of these studies have suggested that ANB leads to a shift in sympathovagal balance towards parasympathetic dominance. Many researchers have found a significant reduction in heart rate (HR) and blood pressure (BP) both after acute and 8 wks ANB[1-3].

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Many studies have shown that both nostrils work alternately in a cyclic manner, not simultaneously. So alternation in nostril breathing is naturally present. Still alternate nostril breathing of yogic type seems to influence parasympathetic nervous system. According to yoga, this phenomenon is a consequence of the alternation of the flow of subtle energy in the Ida and Pingala [2,4-6].

Change in cardiac output and peripheral resistance is very good indicator of change in autonomic status. As they tend to increase with sympathetic stimulation and tend to decrease with increase in parasympathetic activity. Cardiac output is the product of stroke volume and heart rate. Peripheral resistance in the body in man is primarily controlled by the arterioles which are richly supplied with sympathetic fibers, but sparse parasympathetic innervations [7,8].

Cardiac output and peripheral resistance can be measured non invasively by using Impedance Cardiovasograph (Nivomon, L&T Medical's). It is a Non Invasive vasography monitoring system.

It measures the Cardiac Output (CO) and Blood Flow Index (BFI) of the patient non-invasively. It computes the Cardiac Output (CO), Stroke Volume (SV), Systemic Vascular Resistance (SVR), Cardiac Index (CI), Stroke volume Index (SI), Systemic Vascular Resistance Index (SVRI), Pulse Rate (PR) and various other cardiovascular parameters.

Since arterial blood pressure is directly proportional to cardiac output and peripheral resistance. Any maneuver which may decrease cardiac out or peripheral resistance or both may be helpful for the patients suffering from hypertension and other cardiovascular diseases .Therefore, the present study aims the to study the effect of alternate nostril breathing on cardiac output and systemic peripheral resistance.

MATERIAL AND METHOD

The present study was conducted in the department of physiology, Saraswathi Institute of Medical Sciences, Hapur. One hundred asymptomatic healthy male subjects, aged 17-23 years, participated voluntarily in the present study, undertaken, to assess the effect of alternate nostril breathing on cardiac output and peripheral resistance and other cardiovascular parameters. Experiment procedures were in accordance with the ethical committee on human experimentation. Study was carried out at ambient temperature with minimal external or internal sound disturbances in the room. Subjects reported to laboratory 2 hours after light lunch. They were explained in detail about the experimental procedure. Informed consent was taken from all subjects. Subject were asked to lie in supine position. The color coded 8 leads of NICO patient cable were connected at their respective locations as given below:

1. Red leads (I1 and I1′) -Behind the ears (Top pair)
2. Yellow leads (V1 and V1′) -Roof of the neck (Second pair)
3. Violet leads (V2 and V2′) -Level of xiphisternum (Third pair)
4. Green leads (I2 and I2′) End of ribcage or >5 cm from third pair (Bottom pair)

Systolic blood pressure (SBP), diastolic blood pressure (DBP) and heart rate (HR) were recorded by using mercury sphygmomanometer. Cardiac output , peripheral resistance and other parameters were recorded using Impedance Cardiovasograph (Nivomon).

Subjects were asked to assume sitting posture. Then they practiced ANB for 15 min (acute exposure) as per instructions mentioned below

1. Sit in a calm, quiet, airy place in an easy and steady posture with the head, neck and trunk erect and in a straight line and to keep the body still.
2. Bring the right hand up to the nose; fold the index and middle fingers so that the right thumb can close the right nostril and the ring finger can close the left nostril (Vishnu Mudra).
3. With the right nostril closed by the right thumb, exhale completely through the left nostril. The exhalation should be controlled and free from exertion and jerkiness.
4. At the end of the exhalation close the left nostril with the ring finger, open the right nostril and inhale slowly and completely. Inhalation should be smooth, controlled and of the same duration as exhalation.
5. Repeat this cycle of exhalation through the left nostril and inhalation through right nostril, exhale completely through the same nostril keeping the left nostril closed with ring finger.
6. At the end of this exhalation close the right nostril and inhale through the left nostril and repeat this for two more times.

In summary, one exercise consisted of 3 cycles of exhalation through the left nostril and inhalation through the right nostril followed by 3 cycles of exhalation through the right nostril and inhalation through the left nostril and this was repeated for about 15 min.

After 15 minutes ANB exercise, subjects were asked to assume supine position and again all parameters were recorded.

All data were collected and statistical analysis was done by paired t-test using the window SPSS Statistics 17.0 version.

FINDINGS

Table 1: baseline characteristics of all subjects

Data are expressed as Mean±SD.		
S.N.		
1	Age (in years)	20.4±1.2
2	Height (cms)	170.5±2.2
3	Weight (Kg)	65.5±5.3
4	BSA (m^2)	1.76±0.07

Table -2: comparison of cardiac output and peripheral resistance and other cardiovascular parameters before and after alternate nostril breathing (ANB) exercise

S.N.		Before ANB exercise	After ANB exercise
1	Systolic blood pressure (SBP) (mm Hg)	116.2±1.8	106.4±1.2*
2	Diastolic blood pressure (DBP) (mm Hg)	74.12±1.6	66.32±1.5**
3	Heart rate (HR) (per minute)	71.38±0.8	67.4±0.6**
4	Cardiac Output (CO) (L/min)	5.2±0.08	4.8±0.07*
5	Stroke volume (SV) (ml/ beat)	72.84±0.6	71.21±0.5*
6	Systemic Peripheral Resistance (SPR) (dyne.sec/cm^5)	1356.1±8.4	1328±6.4**
7	Cardiac Index (CI) (L/min/m^2)	2.95±0.07	2.72±0.05*
8	Stroke volume Index (SI) (ml/ beat/m^2)	41.39±0.02	40.46±0.03*
9	Systemic Vascular Resistance Index (SVRI) (dyne.sec/cm^5/m^2)	770.5±3.5	754.5±4.1**

*p<0.05 (significant), **p<0.001(highly significant)

Table -2 shows comparison of parameters before and after alternate nostril breathing. There was significant decrease in all cardiovascular parameters after performing 15 minutes ANB. Decrease in Diastolic blood pressure (DBP), heart rate (HR) , Systemic Peripheral Resistance (SPR) and Systemic Vascular Resistance Index (SVRI) were highly significant (p<0.001). while decrease in Systolic blood pressure (SBP), Cardiac Output (CO), Stroke volume (SV), Cardiac Index (CI), Stroke volume Index (SI) were less significant (p<0.05).

CONCLUSION

Blood pressure and heart rate are important cardiovascular parameters. Like other various visceral activities of body, blood pressure and heart rate are controlled by autonomic nervous system. Blood pressure depends on cardiac output and peripheral resistance. The sympathetic nerves that constrict arterioles and veins and increase heart rate and stroke volume, discharge in a tonic fashion, and blood pressure is adjusted by variations in the rate of this tonic discharge [7,8].

Impulses reaching the medulla also affect the heart rate via vagal discharge to the heart. The neurons from which the vagal fibers arise are in the dorsal motor nucleus of the vagus and the nucleus ambiguous.

When vasoconstrictor discharge increases, arteriolar constriction also increases resulting in increase in systemic peripheral resistance and consequently blood pressure rises. Venoconstriction and a decrease in the stores of blood in the venous reservoirs usually accompany these changes, although changes in the capacitance vessels do not always parallel changes in the resistance vessels. Heart rate and stroke volume are increased because of activity in the sympathetic nerves to the heart, and cardiac output is increased [8-10].

Alternate nostril breathing results in decrease in sympathetic activity and increase in parasympathetic activity. As a result of this decrease in sympathetic activity there is vasodilatation which causes decrease in peripheral resistance. It also decreases heart rate and myocardial contractility leading to decreased cardiac output. Decrease in cardiac output and peripheral resistance both results in decrease in systolic as well as diastolic blood pressure. So alternate nostril breathing can be a useful exercise for the patients suffering from hypertension and other cardiac disease and other stress related problems.

Acknowledgement: Nil

Conflict of Interest: Nil

Source of Funding: Nil

Ethical Clearance: Procedures followed in the present study were in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1975, as revised in 2000 (5). Informed concent was obtained from the subjects.

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Irritability Scoring in Working and Non Working Women

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ABSTRACT

Background: Irritability is a prominent symptom in spectrum of female specific mood disorder causing problem to oneself and in relationship with family, friends and community .As there is no precise instrument to measure it , not many studies have been done on irritability especially in females. An attempt has been made to study and score irritability in working and non working females.

Hypothesis: To study the effect of work pressure on working female by scoring their irritability on comparison with non working female.

Material and Method: Study group comprised of 40 working females working at BMCRI establishment office and 40 non working females in Bangalore aged 20 -40yrs.Their irritability was scored using Born Steiner self rating irritability score which has two scales- irritability scoring and visual scoring

Results and Conclusion: Statistical analysis showed that irritability scoring was high in working women compared to non working women .Visual scoring showed that irritability is causing problem for oneself and in relationship with family, friends and community .

Occupation demands and home responsibilities which act as stress inducers, because of which hippocampus, amygdale and prefrontal cortex undergo stress induced remodeling which alters behavioral and physiological responses .leading to high irritability in working females.

Keywords: *Irritability, Stress, Working Female*

INTRODUCTION

Irritability is defined as proneness to anger annoyance or impatience.(1)But many scientists have taken irritability as a mood that predisposes towards certain emotions like anger, certain cognitions like hostile appraisals and certain action like aggression .It is subjectively unpleasant and objectively characterized by expressions of negative emotions in interpersonal relationships. Moods last for days or weeks tends to bias cognitions and may not have specific facial expression unlike emotions which last for seconds to minutes.(2)

Irritability a feature of many psychiatric illnesses like depression and anxiety .As such not treated causes harm to the person and his surroundings. Despite all this there is very little research on irritability.

Working women who are in a competitive world where competition itself acts as a stress. Occupation demands are highly stressful and many jobs make severe demands in terms of responsibility, time and performance. (3)With these additional home responsibilities are equally demanding. All these together are involved in stress induced irritable response which has been linked to other aspects of morbidity like treatment non adherence, suicide attempt and violence.

Hypothesis: To study the effect of work pressure on working female by scoring their irritability on comparison with non working female.

OBJECTIVES

1. To score irritability using Born steiner irritability scale in working women
2. To score irritability using Born steiner irritability scale in non working women
3. To compare scoring between working and non working women

Inclusion criteria

- Age 20-40 yrs
- Cases- female establishment workers
- Controls-home makers
- Regular menstrual history

Exclusion criteria

- Menstrual phase
- Any psychiatric illness
- Any medications
- Any medical illness

METHODOLOGY

Study group consisted of 40 healthy establishment workers female, working at BMCRI establishment office and 40 healthy homemakers of Bangalore city who were age matched considering inclusion and exclusion criteria. General physical examination done to rule out any medical illness and their irritability were scored using Born –Steiner's self rating irritability scale .[4]Subjects were divided according to phase of menstrual cycle as leuteal and follicular phase. Care was taken that they were in the same phase for past 1 week, as the scoring which we are using describes how the subject was feeling past week.

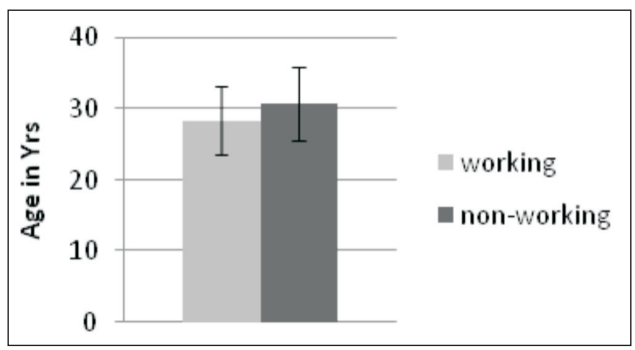
Scale contained sl no 1-14 likerd questions which were specially designed for females and sl no 15-21 visual analogue scale which shows percentage of affection. The scale used is easy to use and interpret.

Sl no 1-14 questions which has scoring for how you are feeling in the past week is answered as not at all, a little or some of time, often and most of the time. They are scored 0, 1, 2 and 3 respectively. Maximum score being 42 and least being 0.

If their score is between 1-14 considered mild irritable,15-28 moderately irritable and 29-42 severe irritable.

Serial no 15-21 have visual scale which has 100mm horizontal line for each criteria .Subject has to mark percentage of affection on that horizontal line, starting point on the line is 0% affection and the end point of line being 100% affection .Later using a ruler scale we can divide the line into 10 segments, each segment represents 10% .Using this we will decide the percentage of affection .This scale shows affection of relationships with family, affection of daily activities, their ability to deal with frustration, affect on their self esteem, affect on their social relationship, how they are feeling at that moment and how they rate themselves.

Results and statistical analysis



Graph 1: Showing age distribution of the subjects (P=0.6)

Table 1: Showing mean irritability scoring of subjects

Score	Working (mean ± SD)	Non Working (mean ± SD)	P value
ISCR (Irritability score)	10.7 ± 6.9	2.97 ± 2.94	< 0.001

Table 2: Showing percentage of subjects with different scoring

	IRRITABILITY SCORING			
SCORE	**0**	**1 TO 14**	**15 TO 28**	**29 TO 42**
RATING	Not irritable	mild irritable	moderate irritable	severe irritable
Working	1(2.5%)	27(67.5%)	11(27.5%)	1(2.5%)
non working	12(30%)	28(70%)	0	0

Table 3: Visual analogue scores showing percentage affect with respect to different criteria for burden of irritability in both groups

Criteria's for burden of irritability	Working (mean ± SD)	Non Working (mean ± SD)	P value
Affect on relationship with family	27 ± 23.6	12 ± 5.1	0.002
Affect on daily activities	20.5 ± 14.7	12.5 ± 6.7	0.002
Affect on ability to deal with frustration	26.5 ± 21.1	13 ± 6.9	0.002
Affect on self esteem	21 ± 15.5	13 ± 9.4	0.006
Affect on social relation	21.5 ± 17.2	13.5 ±10.8	0.01
over all affect of irritability	163.25 ± 81.7	91.02 ± 47.9	< 0.001

RESULTS

Graph 1 shows that cases and controls are age matched with p value 0.6. Table 1 shows mean irritability scoring of working and non working females, which shows working women score to be significantly higher than non working women with P value < 0.001. Table 2 shows Percentage of working and non working women with irritability. As per table 2, 27.5% of working female were moderately irritable and 2.5% were severely irritable. Whereas no one from the non working female group were in this scale.

Table 3 with visual analogue scoring showed irritability has affected working female- relationship with family, their daily activities, their ability to deal with frustration, their self esteem and their social relationship. It also showed that working female rate themselves on higher limit of irritable scale.

DISCUSSION

Occupation demands can be highly stressful and many jobs make severe demands in terms of responsibility, time and performance .With these additional home responsibilities which are stress inducers.

Hippocampus, amygdale and prefrontal cortex undergo stress induced remodeling which alters behavioral and physiological responses. [5] Amino acids and peptidergic neurotransmitter are intricately involved in stress response. Stressors activate noradrenergic system in the brain most notably in locus ceruleus and cause release of catecholamines from the autonomic nervous system. Stressors also activate serotonergic system in brain evidenced by increased serotonin turnover. Glucocoricoid levels increased due to stress, may increase serotonin tryptamine mediated action contributing to intensification of action of these receptor types which has been implicated in pathophysiology of major depressive disorder. Stress also increase dopaminergic neurotransmitter in mesofrontal pathway.[6]

Corticotrophin releasing factor, glutamate ,gamma amino butyric acid all play important role in generating stress response or modulating other stress responsive system such as dopeminergic and nor adrenergic brain activity .Mood is related to amount of norepinephrine available at synapses of brain.

All these together are involved in stress induced irritable response.[7]

Studies especially on irritability is scant .Spyropoulou A et al studied irritability in menopausal women and found it to be related to FSH and LH levels. It was also correlated to depression and anxiety disorder.[8]

Table 3 shows visual analogue scores which highlight the percentage burden of irritability. Shows working women are negatively affected with respect to relation with family, daily activities and social relation (p=<0.01).There ability to deal with frustration

is also negatively affected. They also have significantly less self esteem compared to non working women (p=<0.01).

Working female must cope up with her stress. By understanding the nature of stress and preparing for it, and knowing how long it will last, this will lessen the severity of stress when it comes.

Controlling the stress appears to moderate the effect of stress by allowing the person to alter the stress response directly or to select a response that will alter or divert the threatened event.

Positive social and family relationship can moderate the effect of stress on a person and can reduce its effect. Conversely, lack of support makes stressors more potent and weakens person's capacity to cope with it.

Ultimately what is needed is emotional support which can be expressed by actions as well as words and encourage working female who is doing good by supporting family financially and taking care of family affectionately to do better.

CONCLUSION

Working female managing both family and work is more irritability than non working female.Iriitability is negatively affecting working women relation with family and community.

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Spirometric Changes in Patients with Subclinical Hypothyroidism

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ABSTRACT

Aims and Objective: To study and compare the spirometric changes in patients with subclinical hypothyroid with euthyroid controls.

Material & Method: Thirty diagnosed cases (3 male, 27 female with mean age of 40 ± 7years) of subclinical hypothyroidism (SCH) and thirty age and sex matched healthy controls underwent spirometry test in order to determine if respiratory functions are effected or not. Medical history, physical examination, blood pressure on two different occasion and chest radiogram were performed for all participants. The data of two groups was compared.

Results: The characteristics of SCH patients and control cases were similar with regard to age, sex, and body mass index (BMI). Serum thyroid stimulating hormone levels (TSH) were significantly higher in SCH patients than the controls ($P < 0.001$). There is a significant reduction of spirometric function in all Schs patients when compared to control subjects.

Conclusion: The present study highlights the importance of spirometry test in patients of subclinical hypothyroidism, as an important diagnostic procedure to screen asymptomatic patients for potential respiratory abnormality.

Keywords: *Respiratory Function, Subclinical Hypothyroidism, Spirometry Test*

INTRODUCTION

Subclinical hypothyroid is the term used to describe patients with normal free thyroxine (T_4) and free triiodothyronin (T_3) and TSH levels of more than 5μIU/L, with generally no obvious symptoms of hypothyroidism[1]. It reflects the earliest stage of thyroid dysfunction. It is well known that respiratory functions are affected at thyroid disorders. The ventilatory response to hypoxia was reported to be significantly lower in hypothyroidism[2, 3]. The main factor responsible for hypoxemia and retention of carbon dioxide in hypothyroidism are respiratory muscle weakness, reduction in lung volumes, disturbed ventilation perfusion balance and obesity. Lower levels of thyroid hormones are associated with respiratory problems during sleep, dyspnea at exercise, inspiratory muscle weakness and disturbed diaphragmatic function [4, 5]. since diagnosis of SCH depends on laboratory values, theoretically, no symptoms or signs are expected but still, patients may suffer from somnolence, weakness and fatigue [6, 7]. Muscle strength is also effected in subclinical hypothyroidism. The decrease in muscle strength effects spirometry functions accordingly [8].

In clinical practice, spirometry is the investigation of choice for the overall assessment of pulmonary function and is equated with the PFT in day to day practice. There are not many studies available in literature regarding the influence of subclinical hypothyroidism on respiratory functions. Hence this

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study was conducted with the aim to compare the spirometric values, between SCH patients and euthyroid controls in order to see if these responses vary and can be used as a diagnostic tool to bring out latent respiratory dysfunction.

MATERIALS AND METHOD

Thirty patients, 30- 50 years of age and either sex were selected on the basis of clinical features and biochemical profile (TSH> 5μIU/L, normal free T_3 and $T_{4)}$ suggesting subclinical hypothyroidism. They were recruited from the medical outpatient department, IGIMS Hospital, Patna. After a complete evaluation as described below, they were regularly followed up in the thyroid clinic. The control group comprised of, thirty healthy individuals in same age group as patients, of either sex, with normal T_3, T_4 and TSH. They were selected from staff of hospital, relative of patients and patients with non specific complains attending other clinics. Patients with overt hypothyroidism, hypertension, past history of any respiratory disease, any systemic pathology affecting the respiratory system, pulmonary hypertension, hepatic or renal dysfunction, diabetes mellitus, significant neurological or psychological disease, and those receiving medications such as lithium carbonate, iodine and iodide containing drugs, smokers, pregnant females were excluded. None of the subjects were undergoing regular exercise training in any form.

A complete medical history with physical examination, BP on two different occasions, electrocardiogram, and chest radiogram was done for all participants. The study protocol was approved by the institutional ethical committee, and informed written consent was obtained from all participants prior to study.

For PFT: Medical international research (MIR) Spirolab II (Via Del Maggiolino, 125, 00155 Rome, Italy. 2001) was used.

All the subjects were made familiar with the instrument and the procedure for performing the test. The data of the subjects as regards to name, age, height, weight, sex, date of performing the test, and atmospheric temperature were fed to the computerized MIR Spirolab II.

The tests were performed in sitting position. The subject was asked to take full inspiration which was followed by as much rapid and forceful expiration as possible in the mouthpiece of MIR Spirolab. Three consecutive readings were taken and the best reading amongst the three was selected. We followed the guidelines of American Thoracic Society.

Lung function parameters studied were: forced vital capacity (FVC), FVC%, forced expiratory volume in 1 s (FEV1), FEV1 percentage of FVC in % [FEV1 (%)], FEV1/ FVC, forced expiratory flow rate during 25–75% of expiration (FEF25–75%).

Serum fT3, fT4 levels were assessed by Chemiluminescent Competitive Enzyme Immunoassay method with Access 2 immunoassay system of Beckman coulter. Normal range for TSH was 0.34 -5.00 μIU/ ml, 2.50 – 3.90 pg / ml for fT_3 and 0.61 – 1.12 ng/ dl for fT_4

Statistical analysis

All data are expressed as mean ± standard deviation (SD). The analysis was performed using Graphpad instat prism 6. The chi-square test was used to compare male/ female ratio of SCH patients and euthyroid controls. Statistical significance was accepted at $P<0.05$. Comparison among control and cases were performed by two tailed student's t test. Pearson's two tailed correlation was done between continuous variables.

RESULTS

The demographic characteristics of patients and controls are given in table 1.The mean age and standard deviation of patients was 40.43± 7.182 and it was similar to controls whose age was 41.13± 6.06 ($p = 0.700$). 90% percent of patients and 70% of controls were represented by females and there were no intergroup differences on this basis. All recruited patients were nonsmokers and had a sedentary lifestyle. The TSH of the patients was 7.16. ± 1.23, significantly higher than controls whose value was 3.12 ± 1.06 ($p= 0.000$). None of the patients were treated with thyroxine. The TPO were within normal limit for both groups.

Serum fT3, fT4 and TSH values and spirometric parameters of the groups are shown at Table 2 and 3, respectively.

Comparison between subclinical hypothyroidism and control group showed that all the spirometric parameters were higher in the control group and lower in patients with subclinical hypothyroidism and there

was a statistical significant difference regarding FVC, FVC %, FEV1 % , FEF_{25-75} and FEF25-75 % ($p<0.05$).

There was a positive correlation between fT3 and FVC% (r=0.01385, p=0.9421), FEV1 % (r=0.0479, p=0.8048), FEV1/ FVC (r=0.0479, p=0.8048) and FEF25 -75 (r=0.1077, p=0.5712) in the SCH group. There was also a positive correlation between fT4 and FVC (r=0.1231, p=0.5171) and FEV1 (r=0.2682, p= 0.1519) , FEV1% (r=0.3078, p =0.0979), FEV1 / FVC (r=0.3078, p=0.0979), FEF 25 -75% (r = 0.1162, p =0.5409) FEF 25-75 (r=0.1908, p = 0.3124).

On the other hand, there was a negative correlation between TSH and FVC% (r= -0.249, p=0.02), FEV1% (r = -0.149, p = 0.42). T3 with FVC (r= 0.148, p = 0.334), FEV1 (r =-0.08, p= 0.658), FEF =25- 75% (r = -0.271, p = 0.146). T4 with FVC% (r = -0.003, p= 0.983).

Table 1: Demographic and Metabolic characteristics of patients and controls

Parameters	Subclinical hypothyroid (n = 30)	Euthyroid controls (n= 30)	p value
Age (years)(range)	40. 43 ± 7.82(30 – 50)	41.13 ± 6.06(32 – 50)	0.700
% Females	90%	70%	
Weight (kg)	63.13 ± 6. 32	63. 43 ± 8. 28	0.875
BMI (kg/m^2)	25. 06 ± 2.70	24.62 ± 2. 73	0. 542
Smoking	0 %	0 %	
Sedentary lifestyle	100 %	100 %	
Menopause in female	0 %	0 %	

All results are expressed as Mean ± standard deviation, p< 0.05 is significant

BMI = Basal metabolic index

Table 2: Thyroid function values of patient and control

Parameters	Subclinical hypothyroid (n = 30)	Euthyroid controls (n= 30)	p value
f T_3 (ng/dl)	.279±.078	.303±.069	.216
fT_4 (ng/dl)	1.53±0.44	1.50±0.39	.738
TSH (μIU/L)	7.16± 1.23	3.12±1.06	0.000

All results are expressed as Mean ± standard deviation, p< 0.05 is significant

TSH: Thyroid stimulating hormone, fT_4: free thyroxine, f T_3: free tri iodo thyroxine

Table -3: Spirometric parameters of the patients & controls

Parameters	Subclinical hypothyroid (n = 30)	Euthyroid controls (n= 30)	p value
FVC(ml)	3034.93±242.54	3283.6±537.22	0.0244
FVC (%)	94.68±5.95	111.7±8.18	<0.0001
FEV1(ml)	2787.8±317.86	2786±447.59	0.9876
FEV1 (%)	95.6±6.92	85.06±5.49	<0.0001
FEV1/FVC	91.773 ±6.64	85.057±5.49	<0.001
$FEF_{25-75\ (ml)}$	4628.3±643.04	9298.6±860.97	<0.0001
$FEF_{25-75\%}$	81.17±9.02	89.4±11.14	0.0026

All results are expressed as Mean ± standard deviation, p< 0.05 is significant

DISCUSSION

Subclinical hypothyroidism (SCH), also called mild thyroid failure, is diagnosed when peripheral thyroid hormone levels are within normal reference laboratory range but serum thyroid-stimulating hormone (TSH) levels are mildly elevated.[9] It reflects the earliest stage of thyroid dysfunction. Chronic autoimmune thyroiditis, subacute thyroiditis, thyroidectomy,

treatment with cytokine, lithium, amiodarone and iodine, insufficient thyroid hormone replacement therapy may be the cause of subclinical hypothyroidism.[2, 10]. This condition occurs in 3% to 8% of the general population. It is more common in women than men, and its prevalence increases with age of patients. 80% of SCH patients have a serum TSH of less than 10 mIU/L.

Patients with SCH have a high rate of progression to clinically overt hypothyroidism, 2.6% each year if thyroperoxidase (TPO) antibodies are absent and 4.3% if they are present.[11] However, some persons do not show progression and some experience normalization. A TSH level greater than 10 mIU/L predicts a higher rate of progression, and a level of less than 6 mIU/L predicts a lower likelihood of progression.[12]

The influence of subclinical hypothyroidism on several organ systems are well defined, but very limited study is available showing its effect on respiratory function test. It has been suggested that neuromuscular dysfunction and impaired respiratory function are common in patients with overt hypothyroidism and can be reversed by levothyroxine treatment but its effect on SCH patients is still unclear.[13] Therefore we aimed to evaluate the respiratory function of patients with SCH in comparison with healthy controls. We are using spirometry to assess the respiratory function since this method is simple, cheaper, easily available and non invasive.

In the present study while comparing the lung function between the SCH patients and euthyroid controls, it has been observed a significantly lower values of FVC, FVC %, FEV1 % , FEF_{25-75} and FEF25-75 % ($p<0.05$) in the SCH patients. These results were comparable with the study done by Cakmak et al in which they also found a significant reduction of these parameters in SCH patients when compared with control subjects[14]. Another study by Yilmaz et al also confirmed that subclinical hypothyroidism is associated with decreased vital capacity, FEV1, FVC, and FEF 25- 75%.[15] The possible explanation for these results might be decrease in both expiratory and inspiratory muscle strength16, decrease in maximal breathing and diffusing capacity17and alveolar hypoventilation due to depression of hypoxic and hypercapnoeic ventilatory drives18 in patients with hypothyroidism.

Since Subclinical hypothyroidism reflects the earliest stage of thyroid dysfunction it is hypothesized that the impairment in respiratory function may be initiated at subclinical state. Thus these patients should be routinely screened with simple spirometry test for diagnosis and prevention of further progression to respiratory dysfunction.

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Effects of Examination Stress on Blood Cell Count in First Year Medical Students

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ABSTRACT

Medical students confront with significant academic, psychological, & existential stressors throughout their professional training. Studies have shown that examination stress can have a significant effect on blood cell parameters. The present study was designed to examine the effect of examination stress on different type of blood cells i.e RBC, platelets, haematocrit, TLC, eosinophil, basophil, lymphocyte, monocytes and leucocytes. The changes that occurs are increase in neutrophils and platelets, whereas lymphocytes, eosinophils and monocytes decrease in number.

Keywords: *Examination Stress, RBC, Platelets, Haematocrit, TLC, Eosinophil, Basophil, Lymphocyte, Monocytes, Leucocytes*

INTRODUCTION

Stress is defined as "nonspecific response of the body to any demand, whether it is caused by, or results in, pleasant or unpleasant conditions". Students are subjected to different kinds of stressors, such as the pressure of academics with an obligation to succeed, an uncertain future and difficulties of integrating into the system. There is a considerable amount of stress among medical students and this perceived stress was significantly higher among female students [1]. Many students experience heightened stress levels as examinations approach and in some cases that stress can become very acute, resulting in panic attacks [1]. Medical students confront significant academic, psychological, & existential stressors throughout their training [2]. The students also face social, emotional and physical and family issues which may affect their learning ability and academic performance [2]. Medical student often coping with a new school, new schedule easily feel overextended and as a result students experience some of the symptoms of depression, anxiety & stress [2]. Academic examination stress induces hematological changes that include an increased number of large RBC and increased hemoglobinisation[3]. Stress causes significant increase in the concentrations of cortisol and hemoglobin, and in the values of hematocrit and mean corpuscular volume (MCV) [4]. The stress of examinations in college students produces changes in blood cell parameters and transient rise in systolic blood pressure [5]. These changes include increase in platelets, and neutrophil counts whereas esinophils, lymphocytes and monocytes decreases in number [5]. There are little changes in red blood cell count of both male and female subjects during and before examination [6].

The stress level in medical students increases during academic examination and the label "academic examination stress" covers a wide range of situations that may have very different psychological and immunological consequences [7]. Hence, the results of academic examination studies are often difficult to compare, and conflicting findings are frequent [7].

It is evident from various studies that examination stress can have a significant effect on blood cell parameters. Therefore the present study was designed to examine the effect of examination stress on different type of blood cells.

MATERIAL & METHOD

The present study was conducted in department of Physiology, Santosh medical college and Hospital, Ghaziabad. After initial scrutiny and health checkup, 45 out of 60 shortlisted students of either sex, studying in first year of MBBS, were selected for the final study. The subject for the study were selected based on the following criteria.

Exclusion Criteria

- Subject with high or low blood pressure.(Hypertension > 140/90 mm Hg / Hypotension < 100/60 mm Hg)
- Subjects with high body temperature.(Temp > 100^{o} F)
- Subjects on any long term medication or drugs such as hormones, NSAIDS, vitamins etc.
- Subjects with moderate to very severe case of either depression or stress – scored in DASS-42 questionnaire.
- Heavy Smokers and Alcoholics

Inclusion Criteria

- All healthy first year medical students, who were mentally, physically and medically fit, were included in the study.

Thus selected cohort of 45 1st year medical students were the sample of study, underwent evaluation at two intervals, which is as follows:

1. Pre Examination: Two month prior to start of 1st Professional examination.
2. During Examination: During 1st Professional - Physiology practical examination

A written informed consent was obtained from all the participants. Ethical approval was obtained from Institutional research and ethical committee before starting the study.

Blood Sample Collection

The sample of blood was collected under aseptic conditions at around 9 AM. 1.2 mg of anhydrours salt of E.D.T.A. per milliliter of blood was used as an anticoagulant. Care was taken to avoid frothing of blood during transfer of blood from syringe to the bottle.

Methods for Examination

- RBC, WBC & Platelets : Red Blood, White blood and Platelet examination was performed by haemocytometer.
- Differential leucocyte count (DLC) was done by Glass slides method after staining with Leishman's stain.
- Haematocrit : The haematocrit measurement was performed manually by centrifugation at 10,000 RPM (revolutions per minute) for five minutes.

Statistical Analysis

The result were analyzed by using statistical software Statistical Package of social sciences (SPSS). All values were expressed in mean ± SEM. Student's t-test for paired samples was used to compare the mean values of study group.

RESULTS

Hematological parameters of 45 medical students were examined, before and during Physiology University examination. Hematological parameters included RBC, platelets, haematocrit, TLC, eosinophil, basophil, lymphocyte, monocytes and leucocytes.

Result presented in below mentioned table 1 shows the comparison of various blood cell parameters before and during examination in medical students.

Table 1: Comparison of hematological parameters before and during examination

Parameters	Pre Examination	During Examination	p value
Red Blood Cell (Million/mm^3)	5.1644±0.05	5.1644±0.06	p > 0.05
NEUTROPHIL / mm^3	4495.9±74.35	4584.7±75.55	p < 0.001
EOSINOPHIL /mm^3	221.36±4.34	207.51±4.17	p< 0.001
PLATELETS (Lac/mm^3)	2.69±0.07	3.17±0.07	p < 0.001
MONOCYTES / mm^3	179.93±5.51	189.76±5.01	p < 0.05
LYMPHOCYTES / mm^3	2365.5±37.08	2239.4±38.91	p < 0.001
BASOPHIL / mm^3	24.16±1.92	19.96±1.43	p < 0.01
HAEMATOCRIT (%)	42.89±0.26	42.27±0.28	p > 0.05
Total Leukocyte Count /mm^3	7260.2±117.91	7240.90±120.31	p > 0.05

Values are ± SEM

It is evident from the presented table that red blood cell count changes were non-significant ($p>0.05$) during examination when compared to pre examination with average values of 5.164±0.05 and 5.16±0.06 respectively.

Eosinophil count decreases significantly ($p<0.001$) during examination when compared to pre examination with average values of 207.51±4.17 and 221.36±4.34 respectively.

When average mean value of neutrophil, platelet & monocytes before examination and during examination were compared, highly significant changes ($p<0.001$) were observed in neutrophil count. Result suggests that examination stress increases neutrophil count, platelet & monocytes during examination.

A significant decrease ($p<0.001$) were observed in lymphocyte count, when average mean value of lymphocytes before examination (2365.5±37.08) and during examination (2239.4±38.91) were compared.

When average mean value of basophil before examination and during examination were compared, significant decrease ($p<0.01$) in basophil count were observed.

No significant changes were observed ($p>0.05$) in haematocrit, when results were compared between pre examination and during examination.

TLC count indicated a non-significant changes ($p>0.05$) during examination when compared to pre examination with average values of 7260.2±117.91 and 7240.90±120.31 respectively.

DISCUSSION

Present study employed real life stress situation of acute nature i.e. examination, to show that the stress in first year medical students during examination was significant enough to produce changes in blood cell parameters. Examination Stress is an uneasiness or apprehension experienced during, or after an examination. It is very common among college and university students.

Stress effects the blood cell parameters [2]. Physiological studies have shown that stress from any source can influence on the endocrine, hemopoietic and immune systems [3]. Cytokines and cortisol seem to play an important role in the communication between these systems [3,4]. It has been suggested that endocrine factors released during stress modulate leucocytes trafficking and result in the redistribution of leucocytes between the blood and other immune compartments[8]. The activation of sympathetic nervous system may also have a role to play. Lymphocytes and monocytes express receptors for several stress hormones, including norepinephrine and epinephrine and stressful events alter immune function[8]. This alteration in immune function due to decrease in lymphocytes and basophils was found in the subjects of this study, confirming the stress related changes reported in the study.

The changes that occurs due to stress are increase in neutrophils and platelets, whereas lymphocytes, eosinophils and monocytes decrease in number [4].

The values observed in our study, as shown in the table show significant changes in during-exam values in white blood cell categories. More marked changes were observed in basophil, eosinophil, lymphocyte and monocyte counts. In red blood cell no significant changes were observed and the hematocrit was also not disturbed.

In support of an association between stress and neutrophil activity, stress has been associated with increased numbers of circulating phagocytes and neutrophils bearing activation markers in rats and humans, and this increase appears to be dependent on either or both catecholamines and gluccocorticoids[9].

Examinations in medical schools are particularly stressful as it involves much study and also the results do affect the future study or training of the student. Examinations emphasize the ability to understand, organize and recall the information's. The student is expected to show the depth and breadth of his knowledge. All these can be affected by sheer stress of the situation. Fear of failure or poor performance is quite overwhelming. In this world of competition, unfortunately exams are currently the only means to judge student's knowledge. In the foreseeable future things does not seems to be changing. Students are required to adjust themselves to cope with this stress effectively. The teachers, instructors and other staff members have an important role in behavioral therapy to students. High social support appears to attenuate the magnitude of changes in immune cells suggesting a role for social support in protecting against immune decrements during times of stress[10].

Examinations act as unavoidable stressors, the medical educators as well as students should be made

aware of the negative consequences of stress faced during medical training[11]. Efficient relaxation program as well as counseling services should be provided to stressed students so that they are able to cope better with examination stress[11]. This will enable the students to cope adequately with exam stress and will improve their performances.

CONCLUSION

Medical students confront with significant academic, psychological, & existential stressors throughout their training. Texas Medical Association has defined stress as internal process that occurs when a person is faced with a demand that is perceived to exceed the resources available to effectively respond to it, and where failure to effectively deal with the demand has important and undesirable consequences. Previous studies indicate that stress from any source can influence on the endocrine, hemopoietic and immune systems. Present study was conducted to confirm whether examinations stress in first year medical students was significant enough to produce changes in blood cell parameters.

In this study, conducted in department of Physiology, Santosh Medical College, 45 students of both genders were selected for the study after initial screening and with proper consent. First blood samples were collected from the student 2 months prior to the examination and final blood samples were collected on the day of Practical and Oral examination.

Counting of red blood cells (RBC), total leucocytes count (TLC) and platelets was done by visual means making use of improved Neubauer counting chamber. For differential leucocyte counts (DLC), blood films were stained with Leishman's stain and compound microscope were used to count blood cells. Values obtained during examinations were compared with those taken two month before exams to find out any changes.

The values observed in our study shows significant changes in during-examination values in white blood cell. More marked changes were observed in basophil, eosinophil, lymphocyte and monocyte counts. No significant changes were observed in RBC count and hematocrit percentage.

Blood Cell Parameters	Changes during Examination period
RBC millions/cu.mm.	↔
NEUTROPHIL /cu.mm	↑
EOSINOPHIL /cu.mm	↓
PLATLETS/cu.mm	↑
MONOCYTE /cu.mm	↑
LYMPHOCYTE /cu.mm	↓
HAEMATOCRIT (%)	↔
TOTAL LECUCOCYTE COUNT /cu.mm	↔
BASOPHIL / cu.mm	↓

↑ - Increase, ↓ - Decrease, ↔ - No Change

The present study was limited to show the effect of examination stress on blood cell count only. Moreover study was performed on limited number of subject only. To further elaborate the effect of examination stress, study should be done on large number of subject including students from senior batches of both genders to observe whether results remain same with senior students appearing for 2nd / 3rd Professional examination.

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Effect of Rajayoga Meditation of Brahma Kumaris on Pulmonary Functions

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ABSTRACT

We had measured various parameters of pulmonary functions in 30 healthy medical students, who had never done meditation taken as control and 30 subjects who were practicising rajayoga meditation for more than 2 years regularly taken as a case group. The PFT was measured by computerized spirometer (RMS-Helios, Recorders and Medicare Systems Pvt. Ltd., Chandigarh, India). We have measured Maximum voluntary ventilation (MVV), peak expiratory flow rate (PEFR), forced expiratory volume in the first second (FEV1), forced vital capacity (FVC), forced mid expiratory flow in 25%-75% (FEF25- 75) and FEV1/FVC in who are practicising rajayoga meditation before and after meditation in meditators and compared it with nonmeditators. We found significant changes in FVC, FEF25-75, PEFR in meditators than non meditators.

Highly significant changes in FVC, FEF25-75, PEFR, MVV and significant changes in FEV1/FVC were found after 5 minutes of raja yoga meditation. Results had shown that rajayoga meditations improved the pulmonary functions and thus improve the physical capacity along with providing mental equilibrium and relieving the stress.

Keywords: *Pulmonary Functions Test, Rajayoga Meditation*

INTRODUCTION

Rajayoga practicised by the Brahma Kumaries in which Raja yoga aims at controlling all thought-waves or mental modifications. A raja yogi starts his sadhana with the mind and concentrates the mind and sight between the eyebrows. Rajayoga is the path of Yoga that focuses on meditation and contemplation. Rajyoga meditation of Brahma Kumaris is a behavioural intervention which is simple to practice. Relaxation of body mind with positive approach has been successfully achieved by Rajayoga meditation (autogenic relaxation) providing training in realization of true self. Rajayoga is one of the training courses of Rajayoga Education and Research foundation of Brahma Kumaris World Spiritual University.

Yoga and meditation have been extensively studied for their beneficial effects on human health. The present study is aimed at determining the effect of Raja Yoga Meditation on pulmonary functions. While describing health, W.H.O. in 1948 has defined in the preamble to its constitution that "Health is a state of complete physical, mental & social wellbeing & not mearly an absence of disease or infirmity". Eberst R.M.in 1984 further added spiritual dimension to the concept of health. According to him spiritual health refers to that part of the individual which strives meaning & purpose in life. It is the intangible "something" that transcends Physiology & Psychology. Yoga is one such approach which was widely practiced in ancient India. Yoga influences physical, mental, social, spiritual aspect of human existence.

MATERIAL AND METHOD

We had measured various parameters of pulmonary functions in 30 subjects who were

practicing Raja yoga meditation for more than 2 years regularly taken as a meditators group and 30 healthy medical students, who had never done meditation taken as (control) non meditators. Institutional ethical committee approved this study and written consent was obtained from each subject included in the study. The study was conducted in Dept of Physiology, Saraswathi Institute of Medical Sciences Hapur, Uttar Pradesh.

The Inclusion criteria

We included the Healthy non smoker subject with no cardio respiratory diseases and not doing any other type of exercise.

The exclusion criteria

We excluded the subjects having history of active sports training, previous experience of yoga, history of major medical illness such as tuberculosis, hypertension, diabetes mellitus, bronchial asthma, major surgery in the recent past, smoking, alcohol consumption and non vegetarian diet.

The following Pulmonary function parameters were measured

Maximum voluntary ventilation (MVV), peak expiratory flow rate (PEFR), forced expiratory volume in the first second (FEV1), forced vital capacity (FVC), forced mid expiratory flow in 25%–75% (FEF25- 75).

Minimum three readings were recorded of each test for every subject and the best performed test was selected for precise interpretation of the recorded test.The PFT was measured by computerized spirometer (RMS-Helios, Recorders and Medicare Systems Pvt. Ltd., Chandigarh, India) in comfortable sitting posture.

A] For Forced Capacity measurement: The subject is asked to sit comfortably in an erect posture in a chair. The subject was instructed to take maximum inspiration and blow into the mouthpiece as rapidly, forcefully and completely as possible. It was ensured that a tight seal was maintained between the lips and the mouthpiece of the spirometer. For each parameter, three trials at three minute intervals were given & the best record showing maximum efforts by the subject is selected. FVC, FEV1, FEV1/FVC %, PEFR are recorded simultaneously.

B] For Maximum Ventilatory Volume: The subjects were asked to breath in & out through the mouthpiece as rapidly & as deeply as possible for 12 seconds. After the test is complete the readings of MVV is expressed in Litres/minute. All the measured values are at BTPS (Body Temperature & Pressure Saturated with water vapour). Statistical analysis We have measured FVC, FEV1/FVC, FEF_{25-75}, PEFR, MVV in who are practicing rajayoga meditation before and after meditation in meditators and compared it with non meditators.

Statistical analysis of data The data of both the groups were then statistically assessed using SPSS of windows version 10. Association among variables were assessed using Student's (paired) "t" test for analysis of comparison. P value of less than 0.05 was considered as significant. The data were presented as mean ± standard deviation (SD). Probability value (P) of less than 0.05 was considered statistically significant.

RESULT

We found significant changes in FVC, FEF_{25-75}, PEFR in meditators than non meditators. Which reflect that meditation improves the pulmonary efficiency.

Highly significant changes in FVC, FEF_{25-75}, PEFR, MVV and significant changes in FEV1/FVC were found after 5 minutes of raja yoga meditation, which show that meditation even for short periods can improve the pulmonary functions.

Table 1: Comparison of spirometery parameters in meditators and nonmeditators

	Meditators	Nonmedtators	P-value
FVC (L)	3.24±.54	3.53±.50	.035
FEV1/FVC(%)	86.27±6.23	88.15±7.51	.295
FEF25-75 (L/S)	2.81±.93	2.98±.97	.0001
PEFR (L/S)	5.39±1.09	5.96±1.19	.050
MVV (L/Min)	102.50±28.18	98.73±17.19	.534

Table 2: Comparison of Spirometery parameters after and before meditation in meditators

	Pre Meditation	Post Meditation	P-value
FVC(L)	3.24±.54	3.94±.63	.0001
FEV1/FVC (%)	86.27±6.23	88.15±7.51	.021
FEF25-75 (L/S)	2.81±.93	3.08±.93	.001
PEFR (L/S)	5.39±1.09	5.85±1.36	.001
MVV (L/Min)	102.50±28.18	108.90±27.69	.0001

DISCUSSION

From our results it is evident that the post meditation showed significant improvement in vital capacity and maximal ventilatory ventilation and Peak expiratory flow rate. The findings were supported by

the study conducted by Mauch AD et al[10] , Upadhyay et al[13], Joshi LN, et al[5], Murthy et al[9] in their study reported a statistically significant increase in PEFR.

Joshi et al[5] have reported that pranayam training improves ventilatory functions in the form of increase in FEV, FEV1 and PEFR. Makwana et al[8] and Yadav and Das[14] also found a significant increase in these parameters after yoga training. Thus our results are consistent with the findings of other workers who have reported beneficial effects of yoga training on pulmonary function as measured by spirometry. Vital capacity is a critical component of good health and its determination is important for normal subjects, smokers and patients with respiratory and cardiovascular conditions. PEFR is an inexpensive, accurate and simple test for measuring airway resistance and strength of expiratory muscles.

Our present findings that pulmonary function tests such as FEV, FEV1 and PEFR increased significantly after rajayoga meditation is consistent with earlier studies. Bhole et al [1] have reported a significant increase in vital capacity after three weeks of yoga training. In a study on 287 college students (both men and women), Birkel and Edgren[2] found that yoga training produced a significant improvement in vital capacity across all categories of subjects that included smokers, asthmatics as well as those with no known lung disease

In pranayama there is prolonged inspiration as well as expiration. This stretches the elastin & collagen fibres. Hence after pranayama practice these fibres elongate to a greater extent there by increasing the compliance of the lungs. The surface tension of the fluid lining of the inside walls of the alveoli tends to collapse the alveoli. The surface tension is greatly reduced by surfactant. It is claimed that the lungs inflation near to the total lung capacity which occurs during pranayama is a major Physiological stimulus for release of surfactant hence increase in lung compliance[4]. A study conducted in northern Mexico involving four middle-aged and nine older conventional hatha yoga (CHY) practicing females subjected to 11-week intensive hatha yoga (IHY) program consisting of 5 sessions/week for 90 min (55 sessions) demonstrated improved cardiovascular risk factors (namely maximal O2 consumption -VO2max and high density lipoprotein cholesterol [HDL-C]) in middle-aged and olderwomen.[14]

Also it is observed that yoga practices for long periods cause decrease in oxygen consumption per unit work & blood lactate levels. It indicates better oxygen delivery & improved oxygen utilization due to improved cellular respiration. Hence there is decreased demand for oxygen. As shown by Nagendra & Nagarathna[11], yoga therapy acts by reducing the responsiveness of the tracheo-bronchial tree. As there is decreased responsiveness of the tracheo-bronchial tree there is less frequency of attacks in patients of Bronchial asthma[7].

CONCLUSION

Raja yoga meditation improves the pulmonary efficiency as there is significant increased in FVC, FEF_{25-75}, PEFR in meditators than non meditators.

Meditation improves the pulmonary functions as FVC, FEV1/FVC, FEF_{25-75}, PEFR, MVV all significantly changed after meditation, thus leading to better oxygen supply to the body. Perhaps these effects seem to be due to stress relaxation leading to the better performance of the pulmonary system.

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A Study of Lipid Profile Level in Type 2 Diabetes Mellitus Patients in the Age Group of 35 -70 Years

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ABSTRACT

Patients with diabetes have a 2- to 3-fold higher rate of coronary artery disease (CAD), a 4-fold higher risk of dying during acute myocardial infarction (MI), and 2-fold higher risk of post-MI morbidity than non diabetic persons. Diabetic atherosclerosis, reminded us that this is a heterogeneous condition, and that dyslipidemia is only 1 aspect of the patho physiology. However, dyslipidemia is the most readily measured and, at the present time, the most readily treated aspect of the problem.

In this study 200 patient enrolled , 100 were diabetic and 100 were normally control with age group 35 year to 70yr and result shows There was statistically significant rise in serum total cholesterol (218.11 ± 9.556) in diabetics as compared to Control subjects (194.74 ± 8.105),(t value = 18.651) and (P value = 0.001). There was statistically significant rise in Serum total glycerides (120.56 ± 12.285) in diabetics as compared to Control subjects (106.92 ± 11.686), (t value = 8.045) and (P value = 0.0001).There was statistically significant rise in Fasting Blood Sugar (120.86 ± 9.000) in diabetics as compared to Control subjects (87.97 ± 6.731), (t value = 29.265) and (P value = 0.0001).There was statistically significant rise in LDL (165.41 ± 22.684) in diabetics as compared to Control subjects (101.97 ± 22.917), (t value = 19.674) and (P value = 0.001).There was statistically significant decrease in HDL (43.19 ± 3.421) in diabetics as compared to Control subjects (46.39 ± 3.583), (t value = 6.460) and (P value = 0.0001).

Keywords: *Dyslipidemia, coronary artery disease (CAD), HDL cholesterol, LDL-cholesterol*

INTRODUCTION

Diabetes mellitus type 2, one of the common endocrine disorders characterized by polyuria polydipsia polyphagia and increased susceptibility to infections, commences with insulin resistant which progresses gradually with passage of time. Secondary hyperinsulinism develops to counter it, but it too at one point of time fails to maintain glucose homeostasis resulting in glucose intolerance. The key features of diabetic dyslipidemia are: 1) hypertriglyceridemia, 2) a high proportion of small dense low-density lipoprotein-cholesterol (LDL), 3) low high-density lipoprotein-cholesterol (HDL), and 4) postprandial lipidemia. Plasma LDL levels per se are not usually higher than those of non diabetic patients. Insulin resistance in adipocytes allows exuberant lipolysis stimulated by hormone-sensitive lipase, resulting in excessive free fatty acid (FFA) release into the blood. The excess delivery of FFA to the liver (together with hepatic insulin resistance) results in up regulation of Apo lipoprotein B (apoB), by preventing its degradation. Therefore the liver produces and exports an increased amount of triglyceride. The risk of death from cardiovascular disease (CVD) is from two to six times greater in people with type 2 diabetes than those

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without diabetes and is the leading cause of morbidity and mortality in type2 diabetes. At least 50% of deaths are caused by coronary heart disease (CHD). Although many factors play a part there is considerable evidence that abnormalities in serum lipids and lipid metabolism are important risk factors for this increased incidence of CHD in type 2 diabetes.

AIMS AND OBJECTIVES

To study the effect of age, height and weight on lipid profile level in the age group of 35 – 70 years.

To compare the lipid profile level in type 2 diabetes mellitus and age – matched controls.

MATERIALS AND METHOD

No. of Subject (n=200), Diabetic group = 100, Control group = 100.

Selection of Subjects

Inclusion Criterion

Selection of controls: 100 Subjects with normal fasting blood sugar level were included in control group.

Selection of Diabetics subjects: Total 100 subjects were included in the study and inclusion was based on the levels of fasting blood sugar level Subjects included in the study were in the age range of 35 to 70 years in both the groups .Selected subjects in the study were normotensive .

Exclusion Criteria

Diabetes with complications like neuropathy, retinopathy ischemic heart disease and hypertension. Patients with any concurrent sickness like chronic liver disease or hypothyroidism. Patients on drugs like diuretics and contraceptives etc. Type -1 diabetic patients separated from type -2 by age of onset and dependence on insulin therapy alone to achieve normal blood sugar levels.

(1) Sick admitted patient, (2) having secondary causes of hyperglycemia (3) hyperlipidemia Subjects on lipid lowering drugs (4) Subjects with BMI > 30 kg/ M^2 Smokers and alcoholics.

OBSERVATION& RESULTS

Comparison of anthropometric parameters age and height has been presented in the Table No-1

Table No. 1: Anthropometric parameters in Diabetic and Control group

Parameters	Diabetic Group mean ± SD	Control Mean ± SD	t value	p value	Significance
Age (yr)	47.07 ± 4.105	46.72 ± 4.744	0.524	0.601	Non significant
Height (cm)	164.00±7.909	162.21±12.543	1.310	0.193	Non significant
Weight(Kg)	74.46 ±10.680	73.40 ± 8.763	0.760	0.448	Non significant

Presented data in Table No.1 show that age in diabetic group when compared with control was insignificant. Similarly the table also shows that weight and height were non-significant when results of diabetic group were compared with control group.

Table No. 2: Summary of Fasting blood Sugar (FBS), low density Lipoprotein (LDL) , High Density lipoprotein (HDL), Serum Triglyceride (STG) and Serum Total cholesterol (STC)

Parameters	Diabetics mean ± SD	Control Mean ± SD	t value	p value	Significance
FBS (mg/dl)	120.86± 9.000	87.97± 6.731	29.265	0.0001	Significant
LDL (mg/dl)	165.41± 22.684	101.97 ± 22.917	19.674	0.001	Significant
HDL(mg/dl)	43.19± 3.421	46.39 ± 3.583	6.460	0.0001	Significant
STG(mg/dl)	120.56± 12.285	106.92 ± 11.686	8.045	0.0001	Significant
STC(mg/dl)	218.11 ± 9.556	194.74 ± 8.105	18.651	0.001	Significant

The data in the Table No. 2 shows that Fasting blood sugar levels, LDL and STC were significantly higher in the diabetic group when compared with normal control whereas STG and HDL were significantly lower in the diabetic group.

Table No.3: Percentage rise of Fasting blood Sugar (FBS), low density Lipoprotein (LDL), Serum Triglyceride (STG), and Serum Total cholesterol (STC) in diabetic Group as compared to control group.

Parameters	Diabetics mean ± SD	Control Mean ± SD	% rise in Diabetics to control
FBS (mg/dl)	120.86 ± 9.000	87.97± 6.731	37 %
LDL (mg/dl)	165.41 ± 22.684	101.97 ± 22.917	63 %
STG(mg/dl)	120.56 ± 12.285	106.92 ± 11.686	14 %
STC(mg/dl)	218.11 ± 9.556	194.74 ± 8.105	12 %

To have a clear picture of the obtained data of the present study, percent rise in the fasting blood sugar, low density lipoproteins, serum triglycerides serum cholesterol were calculated in the diabetics and normal control. The percent increase is presented in the Table No. 3.It is evident from the table that the rise in Serum total cholesterol was around 12 %, Serum total glycerides 14 %, low density lipoprotein 63 % and fasting Blood Sugar 37 %, while the decrease in the High density lipoprotein level was 3 % in diabetic Group.

Table No. 4 shows the percent change in the high density lipoprotein levels in diabetics and control group.

Table No.4:- Percentage decrease of High density lipoprotein level in diabetic and control group.

Parameters	Diabetics mean ± SD	Control Mean ± SD	% rise in Diabetics to control
HDL(mg/dl)	43.19 ± 3.421	46.39 ± 3.583	3 %

The data is evident from Table No. 4 that decrease in the High density lipoprotein level was 3 % in diabetic Group when compared with that of values in the control group.

DISCUSSION &CONCLUSIONS

In this study table no 1 shows the comparative evaluation of mean ± SD of Age (yrs) 47.07 ± 4.105 in diabetic and 46.72 ± 4.744 in control group, (t = 0.524 and p= 0.601) which is non-significant, comparative evaluation of mean ± SD of height (cm)164.00 ± 7.909) in diabetic and 162.21 ± 12.543 in Control groups, (t =1.310 and p= 0.193) which is non-significant and comparative evaluation of mean ± SD of weight (Kg)74.46 ± 10.680 in diabetic and 73.40 ± 8.763 in Control groups , (t = 0.760 and p= 0.448) which is non-significant.

The data as evident from table no.1 shows that there is no effect of age, height and weight on the levels of lipid profiles as well as fasting blood sugar when a comparison was done between diabetic and control group.

In this study table no.5 shows mean ± SD of FBS (mg/dl) in diabetics is 120.86 ± 39.000 as compared to control which is 87.97 ± 6.731 gm/dl, t value is 8.288 and p value is 0.000, which is statistically significant. There was a significant rise of the levels of fasting blood sugar in Diabetic Group as compared to control Group.

The table no.6 shows mean ± SD of STG (mg/dl) in diabetics is 120.56 ± 47.285 as compared to control which is 106.92 ± 30.686, t value is 2.428 and p value is 0.017, which is statistically significant. There was a significant rise of the levels of serum total glycerides in Diabetic Group as compared to control Group.

The table no.7 shows mean ± SD of HDL (mg/dl) in diabetics is 43.19 ± 6.421 as compared to control which is 46.39 ± 5.583, t value is 3.694 and p value is 0.000, which is statistically significant. There was a significant decrease observed in the levels of High density lipoprotein in the diabetic Group as compared to control Group.

Observations from table no.8 shows mean ± SD of STC (mg/dl) in diabetics is 218.11 ± 19.556 as compared to control which is 194.74 ± 22.105, t value is 8.341 and p value is 0.000, which is statistically significant. There was a significant rise of the levels of serum total cholesterol in Diabetic Group as compared to control Group.

And table no.9 shows mean ± SD of LDL (mg/dl) in diabetics is 165.41 ± 22.684 as compared to control which is 101.97 ± 22.917, t value is 20.791 and p value is 0.000, which is statistically significant. There was a significant rise of the levels of low density lipoprotein in Diabetic Group as compared to control Group.

In this study table no 10 shows the comparison between control and diabetic Group. There was a significant rise of the levels of serum total cholesterol, serum total glycerides, low density lipoprotein and fasting Blood Sugar as compared in Diabetic Group as compared to control Group, however a significant decrease was observed in the levels of High density lipoprotein in the diabetic Group as compared to control Group.

The rise in Serum total cholesterol was around 12 %, Serum total glycerides 14 %, low density lipoprotein 63 % and fasting Blood Sugar 37 %, while the decrease in the High density lipoprotein level was 3 % in diabetic Group.

Result of the present study showed that All diabetic subjects had significantly higher cholesterol levels (Mean ± SD 218.11 ± 19.556), Triglyceride(Mean ± SD 120.56 ± 47.285) , LDL(Mean ± SD 165.41 ± 22.684), and significantly lower HDL cholesterol (Mean ± SD 43.19 ± 6.421) and FBS(Mean ± SD 120.86±39.000)as compared to Control subjects. In which serum cholesterol was (Mean ± SD(194.74 ± 22.105), (t=8.341, p=0.000),which is statistically highly significant, Serum Triglyceride(Mean ± SD 106.92 ± 30.686) ,(t= 2.428, p= 0.017),again statistically highly significant, LDL(Mean ± SD 101.96 ± 22.917), (t=20.791, p= 0.000) statistically highly significant and significantly higher HDL cholesterol (Mean ± SD(46.39 ± 5.583) ,(t=3.694, p= 0.000) statistically highly significant and FBS (Mean ± SD (87.97±6.731) which is statistically highly significant.

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A Study of Magnesium Supplementation on Motor Nerve Conduction Velocity (N.C.V) in Patients of Diabetic Neuropathy

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ABSTRACT

To study the effects of Magnesium supplementation on motor nerve conduction velocity of common peroneal nerve in patients of diabetic neuropathy.

The study was done in 60 diabetic neuropathy patients attending the neurology O.P.D of S.N Medical college & hospital between February 2009 to August 2010. These patients were divided into two groups. Gp I (n=30) receiving Magnesium and Metformin therapy for a period of 16 weeks. Gp II (n=30) receiving only Metformin therapy.

The blood samples were collected and analysed for S.Magnesium and fasting blood glucose at 0, 4, 8, 16 weeks respectively. Nerve conduction velocity for common peroneal nerve was done at 0, 4, 8, 16 weeks respectively.

Average of males in group 1 was 54.36±1.18yrs and of females was 53.09±1.47yrs and in groupII was 55.94±1.62yrs and of females was 52.83±1.58yrs respectively.

Mean baseline value for N.C.V of Common Peroneal Nerve in group I was 38.08±0.68 and after treatment values at 4 weeks was 39.31±0.61,at 8 weeks 40.58±0.62, at 16 weeks was 44.19±0.77 respectively (P<0.05). Mean baseline value for N.C.V of common peroneal nerve in group II was 37.84±0.75 and after treatment values at 4 weeks was 37.24±0.76,at 8 weeks 38.40±0.76, at 16 weeks was 39.64±0.80 respectively (P>0.05). Stastically significant difference was observed in groupI patients for nerve conduction velocity before and after treatment values at 16 weeks(p<0.05). Stastically insignificant difference was observed in group II patients for nerve conduction velocity before and after treatment values at 16 weeks(p>0.05).

Keywords: *Diabetic neuropathy, Magnesium Supplementation, Motor Nerve Conduction Velocity (Common Peroneal Nerve)*

INTRODUCTION

Diabetes is the leading known cause of neuropathy in the world. Neuropathy is the most common complication and greatest source of morbidity and mortality in diabetes patients. The main risk factor for Diabetic Neuropathy is Hyperglycemia.

Magnesium is known to be necessary for nerve conduction · deficiency is known to cause peripheral neuropathy symptoms and studies suggest that a deficiency in Magnesium may worsen blood glucose control in type 2 diabetes [8]. Oral magnesium supplementation improves both insulin sensitivity and metabolic control in type 2 diabetic subjects with decreased serum magnesium levels [5]. Low serum and intracellular magnesium concentrations are associated with insulin resistance, impaired glucose tolerance, and decreased insulin secretion[1] . Hypomagnesaemia has been linked both to the acute metabolic and late chronic complication of diabetes [2]. A study observed

that the plasma magnesium level has been shown to be inversely related to insulin sensitivity [3]. Insulin-mediated glucose disposal is decreased in normal subjects with relatively low plasma magnesium concentrations[4] . Higher intake of magnesium has been shown to improve glucose and also insulin homeostasis[6] So it would be prudent for physicians who treat diabetic patients to consider magnesium deficiency as a contributing factor in many diabetic complications and as a main factor in exacerbation of the disease itself[7]. Garland HO diabetes mellitus is the most frequent chronic disease associated with secondary magnesium deficit and potential link between the magnesium deficit of diabetes and several diabetic complications like neuropathy[10]. A positive association exists between low serum magnesium (Mg) levels and symptoms characteristic of peripheral neuropathy[11]. Engelen w et.al (2000) demonstrated that under unchanged metabolic control supplementation with could improve nerve conduction[12] .

MATERIAL AND METHOD

This study was carried out in the Department of Physiology and Department of Neurology, S.N. Medical College and Associated Hospitals Agra, over a period of I8 months in Diabetic Neuropathy patients attending the Neurology O.P.D from February 2009 to august 2010.the study patients(n=60) were divided into 2 groups. Group I consists of 30 Diabetic Neuropathy patients receiving Magnesium Supplementation and Metformin therapy for a period of 16 weeks. Group II consists of 30 diabetic neuropathy patients of comparable age and sex receiving only Metformin therapy.

All Patients were subjected to a Detailed History and thorough Clinical Examination specially focusing on Neurological Examination after obtaining his/her informed consent. All patients underwent a detailed Neurologic examination of both the upper and lower extremities. Patients were require to have at least two of the following:

1. Symptoms of Paraesthesia or Dysesthesia.
2. Reduced Vibratory sense below the Knee.
3. Reduced Ankle jerk compared to Knee jerk.
4. Reduced Discrimination and Light Touch sense distally in the Legs.

They were thoroughly examined and investigated for Fasting Blood Sugar, S. Magnesium, Blood urea and s.creatinine at first visit (0 weeks) and subsequently these tests were repeated at 4, 8,16 weeks respectively.

A nerve conduction study (NCS) is a test commonly used to evaluate the function, especially the ability of electrical conduction, of the motor and sensory nerves of the human body.

Motor NCS is performed by electrical stimulation of a peripheral nerve and recording from a muscle supplied by this nerve.

Note: Decrease in nerve conduction velocity depicts demyelination while decrease in amplitude depicts axonal degeneration

Nerve conduction velocity of lower limb - NCV of Common Peroneal Nerve (Motor) of right side only was done on DOS-based two channel NCV/EMG machine (M/S Recorders & Medicare System, Chandigarh, India). motor conduction velocity was measured. For evaluating the level of improvements in the peripheral neuropathy, the type II Diabetic neuropathy patients who were attending O.P.D of neurology clinic S.N Medical College, Agra were divided into two groups:

Group-I: was given Magnesium supplementation (Magnesium chloride sustained release tablets) in the doses of 300 mg/d along with Metformin drug for a period of 16 weeks.

Group-II: was given only Metformin for a period of 16 weeks. They were not given any Magnesium supplementation. Group II patients were of comparable ages were of normal weight for height, on usual diet with no drug taken at the time of examination. Renal and liver function test were normal.

Electrophysiological examination of the tested nerve (Common Peroneal Nerve of right leg) were carried out four times for Group I and Group II patients respectively including the first visit at 0 weeks and the subsequent re-examinations at the end of 4, 8, 16 weeks. Average dose of Metformin was 1.5 g/day.

Inclusion Criteria

1. A known case of diabetes mellitus.
2. Patient showing evidences of diabetes mellitus after being investigated for blood sugar.

3. Patients of diabetes mellitus with sign/symptoms suggestive of diabetic polyneuropathy.

Exclusion Criteria

1. Patients of diabetes mellitus with altered sensorium or disturbed mental state.
2. Patients of diabetes mellitus, having any other diseases known to cause peripheral neuropathy like chronic renal failure, liver failure, hypothyroidism, leprosy, Porphyria etc.
3. Patients with diabetes mellitus, i.e. known to cause peripheral neuropathy like Isoniazid, Phenytoin or who are chronic alcoholics.
4. Patients showing abnormal levels of blood urea, S. Creatinine, Abnormal liver function- tests.

OBSERVATION

Table No. 1. Age and Sex Distribution of Diabetic Neuropathy Patients

Age	Group-I (n=30)				Group II (n=30)				Total I±II (n=60)			
	M	F	Total	%	M	F	Total	%	M	F	Total	%
41-50	4	3	7	23.3	4	5	9	30	8	8	16	26.7
51-60	13	7	20	66.7	9	5	14	46.6	22	12	34	56.7
61-70	2	1	3	10.00	4	2	6	20	6	3	9	15.0
>70	0	0	0	0	1	0	1	3.4	1	0	1	1.6
1963.3%	1136.7%	30	100	18 60%	12 40%	30	100	37	23	60	100	

Table No.l depicts the age and sex wise Distribution of Diabetic neuropathy patients.

In Group I - 66.7% of neuropathy patients belongs to the 51-60 age group.23% belongs to 41-50 age group of patients. No case was found in >70 age group.4 males and 3 females were in the 41-50 age group.13 males and 7 females were in 51-60 age group.2 males and 1 female was in 61-70 age group. Out of 30,19 Males(63.3%) and 11 females(36.7%) were present in group I.

In Group II- 56.7% belongs to 51-60 age group.26.7% belongs to 41-50 age group of Diabetic neuropathy patients.4 males and 5 females were in 41-50 age group.9 males and 5 females were in 51-60 age group.4 males and 2 females were in 61-70 age group. 1 male was present in > 70 age group. Out of 30.18 males (60%) and 12 females (40%) were present in group II. In TOTAL-out of 60 diabetic patients 37 were males (61.7%), 23 were females (38.4%)

Table No. 2. Motor nerve conduction velocity of CPN (m/s) before and after treatment

	Group I (n=30)				Group II (n=30)			
	Before treatment	After Treatment			Before treatment	After Treatment		
		4 weeks	8 weeks	16 weeks		4 weeks	8 weeks	16 weeks
Mean	38.08	39.31	40.58	44.19	37.84	37.24	38.40	39.64
S.D	3.72	3.33	3.37	4.21	4.11	4.15	4.18	4.36
S.E.M	0.68	0.61	0.62	0.77	0.75	0.76	0.76	0.80
% Change				16%				4.8%
Over Baseline p Value				P<0.05				p>0.05

Table No. 2 depicts the baseline values (before treatment) of motor N.C.V (m/s) of C.P.N and after treatment values for Group I and GROUP II at 4 weeks, 8 weeks, 16 weeks respectively.

In GROUP I mean Baseline value for motor N.C.V of C.P.N is 38.08±0.68 and after treatment values at 4 weeks is 39.31±0.61,8 weeks 40.58±0.62, at 16 weeks 44.19±0.77 respectively. The difference in this group was highly significant (p<0.05)

In GROUP II mean Baseline value for motor N.C.V of C.P.N is 37.84±0.75 and after treatment values at 4 weeks is 37.24±0.76, 8 weeks 38.40±0.76, at 16 weeks

39.64±0.80 respectively. The difference in this group was not significant (p>0.05).

DISCUSSION

Average age of males was 54.36±1.18 years and of females was 53.09±1.47 year in group I. In group II Average age of males was 55.94±1.62 years and of females was 52.83+1.58 years. Mean duration of diabetes in group I was 15.516±1.066 as compared to 15.290±1.237 in group .A commonly cited study in 1977 reported that roughly 7% of patients had neuropathy upon diagnosis of diabetes, and the incidence approached 50% for patients with diabetes for more than 25 years.

In present study mean value for motor NCV of C.P.N in group I before treatment was 38.08+0.68 m/sec. as compared to 44.19±0.77 m/sec. after treatment (i.e after 16 weeks). This Difference was considered extremely significant (p<0.05). In group II before treatment (i.e first visit) Mean value for motor NCV of C.P.N was 37.84±0.75 m/sec. as compared to 39.64±0.80 m/sec. after treatment (i.e after 16 weeks). This difference was statistically insignificant (p>0.05).

Our results are in accordance with the findings of following studies

Perkins BA *et .al.* (2001) concluded in their study that Glycemic control is related to the morphological severity of diabetic Sensorimotor Polyneuropathy (DSP) as determined by fiber density (FD) on Sural nerve biopsy[9]. Ivo De Leeuw *et. al.* (2004) observed that long term Magnesium supplementation influences favourably the natural evolution of neuropathy in Mg depleted type 1 diabetic patients (Tldm) Chronic Mg depletion in Tldm has been linked to Polyneuropathy (PNP[13]. Engelen W *et. al.* (2000) conducted a study which demonstrated that under unchanged metabolic control, supplementation with Magnesium could improve nerve conduction[12]. Elamin A *et. al.* (1990) observed that appropriate Magnesium supplementation might prove beneficial in normalizing the low plasma and tissue Magnesium levels and prevent or retard the development of vascular & neural complications in diabetic patients[2]. Sophie Begon *et .al.* (2000) observed that Magnesium could be an alternative for the treatment of neuropathic pain in patients of diabetic neuropathy[14].

CONCLUSION

Thus it can be concluded from the present study that Magnesium supplementation along with regular anti-diabetic therapy improves nerve conduction velocity and may help in the improvement of the symptoms of diabetic neuropathy patients. A major limitation of our study was relatively small sample size with diabetic neuropathy patients,So further study involving large number of diabetic neuropathy patients should be done.

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Baroreceptor and Chemoreceptor Reflex Contribution in Cardiovascular Changes during Exercise

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ABSTRACT

During exercise, both baroreceptors and chemoreceptors contribute in bringing about the cardiovascular changes, but less is documented about the contribution made by either of these mechanisms. The purpose of the present study was to demonstrate the differential effects of baroreceptors and chemoreceptors on cardiovascular parameters during exercise. Twelve healthy individuals were subjected to exercise maneuver on stationary cycling machine. Systolic blood pressure (SBP), diastolic blood pressure (DBP) and electrocardiogram (ECG) were recorded during rest and immediately after exercise. Heart rate (HR) and heart rate variability (HRV) was computed from ECG. SBP, DBP, mean arterial blood pressure (MABP) and HR significantly increased due to exercise. Increase in SBP, DBP, Pulse pressure (PP), MABP and HR were more pronounced when chemoreceptor influence was not considered as against when changes due to both baroreceptors and chemoreceptors were considered. The HRV analysis reflected increase in the low-frequency (LF) power % and decrease in high-frequency (HF) power % along with reduction in LF/HF ratio due to exercise, but these changes were statistically not significant. Baroreceptors through baroreflex mechanism overall increase the CVS parameters. The novel finding in this study was the opposing action of chemoreceptor mechanism to that of baroreceptor mechanism in cardiovascular homeostasis during exercise.

Keywords: *Baroreceptors, Chemoreceptors, Exercise, HRV Analysis*

INTRODUCTION

Nervous pathways of the autonomic nervous system (ANS) form the components of baroreceptor and chemoreceptor reflex loops regulating the cardiovascular system (CVS) homeostasis. Recent studies show that the variables which reflect the complex interaction between the ANS and the CVS during exercise testing can provide significant prognostic information.[1-3] Specific variables of importance include blood pressure (BP) at rest and in response to exercise; heart rate (HR) at rest and in response to exercise; and heart rate variability (HRV) both at rest and with exercise. Poor HR response to exercise has been strongly associated with sudden deaths.[4] HRV components give an insight into the autonomic balance between parasympathetic and sympathetic influences on the CVS.

Exercise increases the tissue oxygen demand whereby oxygen homeostasis at the tissue and cellular level occurs by acute and appropriate cardiovascular adjustments brought about by baro- and

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chemoreceptor reflexes. Peripheral chemoreceptor (carotid and aortic bodies) activation cause sympathetic excitation which concurrently modifies the cardiovascular and respiratory responses. Moreover changes in sensitivity of baroreceptor reflex control during chemoreceptor activation may determine the overall short-term nervous mediated cardiovascular regulation. This collective information suggests that these interactions may contribute importantly to cardiovascular pathology requiring considerable research into the linkage between cellular mechanisms to evolve integrated systems approach for understanding CVS responses in health and disease.[5] Changes in ANS balance can be demonstrated by evaluating several discrete aspects of the CVS on exercise test. During exercise, both baroreceptors and chemoreceptors contribute in bringing about the cardiovascular changes, but less is documented about the contribution made by either of the mechanisms. The aim of our study was to document the differential contribution by baroreceptors and chemoreceptors during exercise.

MATERIALS AND METHOD

A study on twelve apparently healthy volunteers in the age group of 16 – 35 years belonging to both genders was taken up after taking written informed consent and approval by Institutional Ethics Committee. Systolic BP (SBP) and diastolic BP (DBP) were recorded using stethoscope (Microtone Co., India) and sphygmomanometer (Diamond Co., India). Pulse pressure (PP) and mean arterial BP (MABP) were then computed mathematically (PP=SBP+DBP, MABP=DBP+ ⅓ PP). Lead II ECG was recorded using Digital Polygraph – RMS POLYRITE-D (Recorders and Medicare Systems Pvt. Ltd., India). HR and HRV components were calculated form ECG by inbuilt software. HRV analysis was done using Fast Fourier Transform (FFT) spectrum through Welsch's periodogram method.

Individuals were subjected to exercise on a stationary cycling machine (Bicycle ergograph, Sayani's Surgicals, India). Cycling predominantly involves activity of lower limb musculature with only minimal contribution by the rest of the muscles in the body. The Riva-Rocci cuffs (23cm X 12.5cm) were tied on the upper aspect of thighs and inflated to 80 mm Hg during exercise which majorly inhibits the venous return without compromising the arterial supply (though some flow occur in deep veins). The cuff tied to the thighs impedes the movement of metabolites from the exercising muscles of lower extremities to reach the carotid and aortic bodies thereby obtunding the chemoreceptor mediated CVS response.

The BP and ECG of the subjects were recorded; HR and HRV noted in various phases of exercise protocol (Table 1).

Table 1: Exercise protocol

Phase	Activity
1	Basal (resting) level
2	Pedaling at moderate velocity. At the end of 10 min HR, ECG, HRV and BP were noted.
3	Adequate rest for 10 min after phase 2 exercise.
4	Pedaling at moderate velocity with thigh cuffs inflated to 80 mm Hg. At the end of 10 min CVS parameters were recorded again.

Difference of values between phase 2 and phase 1 is the collective measure of CVS changes due to both baroreceptor and chemoreceptor reflex actions. Changes due to almost exclusive baroreceptor action can be known by calculating the difference between phase 4 and phase 1. Difference between phase 2 and phase 4 gives almost exclusive contribution of chemoreceptors on CVS changes during exercise.

Statistical analysis

Statistical analysis was performed using SPSS version 17.0 (SPSS Inc., Chicago, IL, USA) and Origin Pro 8.0 (Origin Lab, Northampton, MA, USA). Repeated measure ANOVA was used to analyze the differences between the normally distributed data where as Friedman non-parametric test was used for the data which did not follow normal distribution. The p value<0.05 was considered significant.

RESULTS

Table 2: Test parameters

Parameter	Resting (Phase 1) [Mean ± SD]	After exercise without cuff inflation (Phase 2) [Mean ± SD]	After exercise with cuff inflation (Phase 4) [Mean ± SD]	Change due to contribution of both baroreceptors and chemoreceptors (Phase 2-Phase 1)	Change due to contribution of only baroreceptors (Phase 4-Phase 1)	Change due to contribution of only chemoreceptors (Phase 2-Phase 4)
BP(mm Hg)						
SBP	112.50±9.18	124.50±11.54	129.75±15.27	12.00	17.25	-5.25
DBP	78.60±7.96	86.83±8.09	88.08±6.21	8.23	9.48	-1.25
PP	33.83±6.23	37.66±6.66	41.66±13.37	3.83	7.83	-4.00
MABP	89.94±7.86	99.38±8.84	101.97±7.99	9.44	12.03	-2.59
HR (beats/min)	74.00±11.81	86.91±14.49	89.66±17.48	12.91	15.66	-2.75
HRV(power %)						
LF	60.10±35.78	71.85±32.54	80.23±24.85	11.75	20.13	-8.38
HF	14.11±13.00	9.50±8.49	12.22±9.85	-4.61	-1.89	-2.72
VLF	25.78±29.34	18.63±33.97	7.54±26.12	-7.15	-18.24	11.09
LF/HF	103.18±285.91	36.00±93.48	91.35±285.90	-67.18	-11.83	-55.35

Negative sign (-) indicates decrement

Table 2 shows various cardiovascular parameters during different phases of exercise protocol. SBP and DBP increased during exercise both in phase 2 and phase 4 compared to phase 1. A repeated measures ANOVA with a Greenhouse-Geisser correction determined that mean SBP differed statistically significantly between different phases of exercise protocol ($F(1.329, 14.618) = 21.379$, $p<0.001$). Post hoc tests using the Bonferroni correction revealed that exercise significantly increased SBP in phase 2 ($p<0.001$) and 4 ($p=0.001$) compared with phase 1. However, the change in SBP from phase 2 to phase 4 was not significant ($p=0.178$). A repeated measures ANOVA determined that mean DBP differed statistically significantly between different phases of exercise protocol ($F(2, 22) = 10.353$, $p=0.001$). Post hoc tests using the Bonferroni correction revealed that exercise significantly increased DBP in phase 2 ($p<0.001$) and 4 ($p=0.015$) compared with phase 1 but the change from phase 2 to phase 4 was not significant ($p>0.05$). In effect, there was a predominant increase in SBP than DBP by the activity of baroreceptor reflex whereas chemoreceptor reflex slightly decreased both SBP and DBP (Table 2).

A repeated measures ANOVA with a Greenhouse-Geisser correction determined that mean PP differed statistically significantly between different phases of exercise protocol ($F(1.190, 13.091) = 4.528$, $p=0.04$). Post hoc tests using the Bonferroni correction revealed that PP significantly increased from phase 1 to phase 2 ($p=0.01$) whereas differences between phase 1 and 3; and phase 2 and 3 were not significant ($p>0.05$). A repeated measures ANOVA determined that MABP differed statistically significantly between different phases of exercise protocol ($F(2, 22) = 18.643$, $p<0.001$). Post hoc tests using the Bonferroni correction revealed that MABP was significantly elevated in phase 2 ($p<0.001$) and phase 4 ($p=0.002$) from phase 1 while the rise from phase 2 to 3 was not significant ($p>0.05$). Therefore, we can conclude that baroreceptor reflex has incremental effect and chemoreceptor reflex has decremental effect on PP and MABP.

A repeated measures ANOVA determined that mean HR differed statistically significantly between different phases of exercise protocol ($F(2, 22) = 11.852$, $p<0.001$). Post hoc tests using the Bonferroni correction revealed that HR significantly increased in phase 2 ($p=0.005$) and phase 4 ($p=0.008$) from phase 1 while the increase from phase 2 to 3 was not significant ($p>0.05$). Hence, from table 2 it can be inferred that baroreceptor mechanism mainly elevated HR while the chemoreceptor mechanism showed lowering effect.

HRV components showed equivocal results. Results in table 2 show that low frequency (LF) component increased during exercise whereas very low frequency (VLF) and high frequency (HF) component were reduced. Decrease in LF/HF ratio was drastic in phase 2 but relatively less in phase 4. However, Friedman test to know the statistical significance for these HRV changes revealed no significant Chi-square values ($p>0.05$).

DISCUSSION

Baroreceptor and chemoreceptor reflexes exert considerable influence on autonomic control of the heart and blood vessels, especially in stress situations such as marked changes in BP, partial pressures of oxygen (PaO_2) and carbon dioxide ($PaCO_2$) in arterial blood. The role of baroreflexes and their responses to BP alterations; and of chemoreflexes and their responses to hypoxia and hypercapnia have been previously studied.[6]

In our study, we found that SBP, DBP, PP and MABP were raised to a higher degree when chemoreceptor response was blunted as compared to rise when both baroreceptors and chemoreceptors activity was at its optimum. One study has demonstrated that vasoconstrictor responses to peripheral chemoreceptor stimulation were inhibited by elevation of BP and activation of baroreceptor reflexes.[7] The sympathetic vasoconstriction in exercising skeletal muscles may be attenuated by sympathetic vasodilator fibers and local factors like potassium, lactic acid, etc. which directly relaxes the vascular smooth muscles.[8] However, the interaction of this sympatholytic event with reflex arterial BP control mechanisms remains unclear. Few studies[9-11] have considered the influence of metabolic factors on sympathetic control of the skeletal muscle vasculature during exercise. In the present study, the decremental effect of lone chemoreceptor activity on BP also strengthens the notion that the metabolites produced as a result of exercise have a predominant local effect, thereby decreasing vascular resistance and hence BP. The importance of local metabolic events on arterial BP regulation is accentuated by the fact that increase in blood flow to the exercising muscle changes the vascular conductance in the skeletal muscle producing more pronounced systemic effects.[12,13] Also inflation of cuffs increases the venous resistance. Since the rise was noticed in both SBP and DBP, therefore PP rise was relatively small during exercise. Thus, effective neural reflex and local metabolic control of the blood vessels in exercising muscle is important to ensure that adequate tissue perfusion is achieved without compromising on systemic arterial pressure. Studies have shown that sino-aortic reflex control of arterial BP (baroreceptor mechanism) is effective from rest to exercise.[14,15]

Present study revealed that the change in HR was more when chemoreceptor activity was largely obtunded by impeding the flow of metabolites to the carotid and aortic bodies compared to the change when both baroreceptors and chemoreceptors were working optimally. One important factor to be considered is that an acute episode of exercise predominantly stimulates sympathetic trunk and does not increase vagal tone. The rise in HR found in this study also confirms the activation of sympathetic component of autonomic nervous system. The negative modulatory effect of chemoreceptor reflex mechanism on HR was minimal.

Studies[16-19] on HRV during exercise seemed to indicate that HF power decreases but we found increase in power percentage, though the increase was not statistically significant. The LF component increased during exercise in our study which is in agreement with the findings of Wrey et al[20] but contradictory to other studies.[16-19] Exercise testing has shown that HRV does not exhibit the expected LF/HF ratio, consistent with sympathetic hyperactivity during exercise and parasympathetic drive during recovery. Our results demonstrated decrease in the VLF component and LF/HF ratio during exercise. These findings suggest that HRV is probably best explained by the complex interplay of multiple inputs to the heart, rather than simply autonomic imbalance.[21-22]

CONCLUSIONS

This study aimed at addressing the integrated physiology of cardiovascular modulation during muscular exercise. Baroreceptors through baroreflex mechanism appear to overall increase the CVS parameters. The novel finding in this study was the opposing action of peripheral chemoreceptor mechanism to that of baroreceptor mechanism in cardiovascular homeostasis during exercise. Future studies may improve the utility of the exercise testing in sports medicine and in risk analysis of cardiovascular patients.

Limitations

There are few limitations in our study. Firstly, the subjects were less. Secondly, the role of psychic stimuli in changing the CVS parameters by the thought of the exercise was not accounted. Thirdly, the actions of the muscles in the upper part of the thigh and around the pelvic girdle and also subtle movements of the other parts of the body were not considered for practical reasons. Fourthly, flow in deep veins during inflation of cuff was considered negligible.

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Effect of Thyroidectomy and Thyroxine on Glucose Transport in the Everted Small Intestine of Rat

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ABSTRACT

Thyroid hormone has been shown to alter glucose metabolism, however, there seems to be controversial reports on the effect of thyroxine on intestinal glucose transport. This study was conducted to investigate the effect of thyroidectomy and thyroxine on glucose transport in the small intestine. Forty rats were randomly selected into four groups of ten rats each. Groups one and two rats were thyroidectomised to make them hypothyroid after which group two rats were given thyroxine replacement of 10ug/100g b/w for thirty five days to make them euthyroid. Rats in groups three and four were sham operated thereafter group three rats were given 10ug/100g b/w thyroxine for thirty five days to make them hyperthyroid. Group four served as the control. 10mg/kg b/w Ketamine was administered intraperitoneally as anesthesia for the surgeries. On the thirty-fifth day post-surgery all the animals were killed via cervical dislocation and their small intestines were harvested. 10cm length of jejunum and ileum respectively were used to make everted sacs for the invitro study. Mucosa glucose transfer (MGT), Final glucose concentration gradient (FCG) and Gut glucose uptake (GGU) were significantly higher ($P< 0.05$) in the hyperthyroid group and lower ($P< 0.05$) in the hypothyroid group compared with the control with transport in the jejunum greater ($P< 0.05$) than the ileum in all groups. Serosal glucose transfer (SGT) was Negative in the hyperthyroid group. SGT was significantly greater ($P< 0.05$) in the jejunum than the ileum. These findings suggest that thyroidectomy causes reduced glucose transport while thyroxine increases glucose transport in different segments of the small intestine with the transport in the jejunum greater than that of the ileum.

Keywords: *Glucose, Transport, Thyroxine, Thyroidectomy, Everted Sac*

INTRODUCTION

Thyroid dysfunction is one of the most common endocrine disorders and it appears to be linked with Diabetes Mellitus (DM) with a mean frequency of thyroid disease of 11% in DM patients[1]. Thyroid hormones have well-described effects on glucose metabolism both by short-term and long-term interaction with the regulatory network for glucose homeostasis. However glucose regulation begins with intestinal absorption or transport.

In the past there have been reported studies on intestinal glucose transport under altered thyroid states. Some scientists reported that in rats, removal of the thyroid and the parathyroid glands do not produce any severe disturbance of carbohydrate metabolism. Halliday reported that glucose transfer across the isolated everted intestine of mice was inhibited when the animals were fed with 0.5 % desiccated thyroid for 14 days, Levin and Smyth observed that hyperthyroid rats show little change in their hexose transfer mechanism across the isolated intestine[2,3]. These reports seem controversial thus this study was designed to investigate into the effect of thyroidectomy and thyroxine on glucose transport in different segments of the small intestine.

MATERIALS AND METHOD

Experimental Animals: Sprague Dawley albino rats (100-180g) were obtained from the Central Animal house of the College of Medicine, University of Ibadan, Nigeria. The animals were maintained under standard environmental conditions and provided standard food

pellets and clean water *ad libitum.* Experimental protocols complied with the 'Principle of Laboratory Animal Care.'

The rats were grouped into four of ten rats each. Rats in the first group were thyroidectomised and kept for 35 days to serve as the hypothyroid group, the second group was thyroidectomised and given 10ug/100g bd wt levothyroxine (Forley Generics Ltd, UK) replacement, orally, for 35 days to serve as the Euthyroid group. The third group was sham operated and given 10ug/100g body weight of levothyroxine orally for 35 days to serve as the hyperthyroid group. While the fourth group was sham operated only and served as the control. Thyroidectomy was surgically done and sham operation after anesthesizing the rats with 10mg/kg b/w Ketamine administered intraperitoneally

Thyroxine (T4) Assay: On the thirty fifth day post surgery blood was collected via cardiac puncture from the rats. The blood was centrifuged at 4000rpm for 30 minutes to separate the serum. T4 level was determined from the serum using chemi-immunoluminiscence.

The everted sac study

The rats were killed on the thirty fifth day post surgery by cervical dislocation. The abdomen was opened by a midline incision; the small intestine was removed by cutting across the upper end of the duodenum and the lower end of the ileum at the ileoceacal junction. The intestine was everted using a stainless steel rod to push the ileal end of the gut into the gut lumen until it appeared at the duodenal opening of the intestine, and rolling the proximal half of the intestine over the rod. The eversion exposes the highly active mucosa to the oxygenated suspending medium with the mucosa on the outside and the serosal now on the inside. About10 cm length of everted jejunum or ileum was cut and tied off at one end by a thread ligature. At the other end a blunt needle, attached to a syringe was introduced into the intestinal lumen and a loose ligature pulled tight over the needle. 2-3 mls of Krebs bicarbonate solution was injected into the sac, the needle was withdrawn and the ligature tied tight thus distending the sac. The distension increases the surface area of the sac and reduces the thickness of the sac wall while the fluid in the sac served as the serosal fluid. The distended sac was incubateded in a mucosal fluid of 15ml of krebs bicarbonate solution for 30mins after which the sac was removed, opened and samples of the serosal and mucosal fluids were taken for estimation of glucose concentration[4,5]. Glucose concentration was determined by glucose oxidase/peroxidase technique using accucheck glucometer.

The initial and final mucosal and serosal concentrations is the glucose concentration in the mucosal and serosal fluid at the beginning and end of the experiment respectively; the final concentration gradient (FCG) is the difference between the final serosal glucose concentration (FSC) and the final mucosal glucose concentration (FMC); the mucosal glucose transfer (MGT) is the amount of glucose which disappears from the mucosal fluid; the serosal glucose transfer (SGT) is the increase in glucose concentration in the serosal fluid; the gut glucose uptake (GGU) is the difference between MGT and SGT. These quantities as defined have been considered positive however SGT may be either increased (positive) or decreased (negative). Positive FCG implies that FSC was higher than the FMC. Glucose transfer was expressed as mg/g sac/30 minutes[6].

Statistical Analysis

Results were expressed as mean ± S.E.M. The statistical significance of differences was estimated with GraphPad Prism version 4.0 using "Newman-Keuls Multiple Comparison Test ANOVA". $P < 0.05$ was considered to be significant.

RESULTS

Table 1 Shows the concentration of T4 in the rats before the experiment.

Table 1: Thyroid hormone concentration in the rats on the 35th day post-surgery

GROUP	T4 (nmol/L)
Control	30.4
Hypothyroid	17.4**
Hyperthyroid	49***
Euthyroid	27.1[a]

The Values are expressed as mean ± S.E.M. (n = 10 rats). **P<0.01, ***P<0.001 significant difference, aP> 0.05 no significant difference

Transfer of glucose was studied by taking nine variables from the same set of experiment. Each set of experiment was carried out on sacs made from both the jejunum and ileum. Difference in glucose concentration was observed due to the transport activities of glucose across the sacs (Table 2)

Mucosal glucose transfer: There was significant loss of glucose from the mucosal fluid with greater loss in the jejunum of the hyperthyroid group. Percentage mucosal glucose transfer was highest (P<0.001) in the hyperthyroid jejunum and ileum and least in the hypothyroid with transfer in the jejunum greater than the ileum in all the groups (Table 2).

Gut glucose uptake (GGU): A large part of the mucosal transfer is both retained in the gut wall and metabolized. This is referred to as gut glucose uptake. GGU is highest in hyperthyroid jejunum (Table 2)

Final concentration gradient (FCG) was positive and greater in the jejunum than ileum across all the groups. It was greatest in hyperthyroid and least in hypothyroid groups (Table 2).

Table 2: Glucose transfer capacity of different segments of the small intestine in different thyroid states.

	Group Control		Hypothyroid		Hyperthyroid		Euthyroid	
Description	**J**	**I**	**J**	**I**	**J**	**I**	**J**	**I**
Initial mucosal & serosal conc (mg/dl)	522	522	522	522	522	522	522	522
FMC(mg/dl)	468.4±13	489.0± 6.7	494.4±6.8	501±2.9	400.8± 11.6	447.2 ± 4.2	463±8.4	475.4± 2.8
FSC(mg/dl)	544.2±4.3	534.8± 1.7	543±6.8	541.8±3.5	497.4±2.4	501.8 ± 2.5	529.8± 5.8	518.6 ±5.5
FCG(mg/dl)	75.8±16	45.2±7.5	48.6 ±6	40.8±2	96.6± 12.5	54.6 ±5	66.8 ±9	43.2 ±5.8
MGT(mg/g)	67±16.4	40.5 ±8	21.2±5	16.8±2	151.5±14.5	93.5±5	62.1±8.8	58.25±3.5
SGT(mg/g)	27.8±5.4	16±2.1	16.15±5.3	15±2.8	--30.8±3	--25.3± 3.1	8.2±6	--4.25±6.9
GGU(mg/g)	39.3±13.7	24.5 ±7.8	5.08± 9	1.09±5.5	182.3± 14	118.75±5	53.8±12	62.5±8.2
Mucosa glucose absorbed(mg/ cm/30mins)	6.7 ±1.6	4.05±0.8	3.3±0.9	2.1± 0.3	12.12±1.2	7.48±0.4	5.9±0.8	4.66±0.3
Serosa glucose absorbed mg/ cm/30mins	2.8 ± 0.5	1.6 ±0.2	2.1±0.7	2.5± 0.4	-2.46± 0.2	-2.02± 0.25	0.8±0.6	-0.34± 0.5

j=jejunum, i-ileum, n=6, mean ± *SEM*.

Serosal glucose transfer (SGT): There was a negative serosal glucose transfer in the hyperthyroid group and Euthyroid ileum. Serosa glucose absorbed (SGA) is negative in the hyperthyroid group (-2.46± 0.24 mg/cm/30mins) but positive in the other groups (Table 2). This implies that some glucose actually disappears from the serosal fluid. % SGT is greatest in the hyperthyroid group but in the reverse direction (Figure 2).

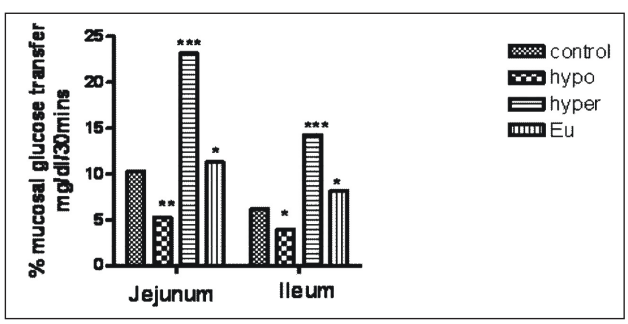

Fig. 1. Percentage Mucosa glucose transfer across the isolated jejunum and ileum of thyroidectomised and thyroxine treated rats

percentage mean,n=6, *P<0.05,**p<0.01,***P<0.001

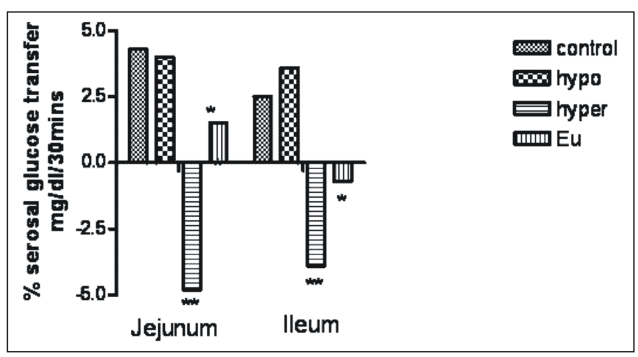

Fig. 2. Percentage serosal glucose transfer

Values are expressed as mean ± S.E.M. (n = 6 rats). *P<0.05, **P<0.01 significantly different

DISCUSSION

Treatment efficacy was confirmed by determination of serum T4 level to ascertain the establishment of the altered thyroid states in the rats. The hypothyroid group has a significantly lower serum T4 level than the control 35days after the removal of the thyroid gland thus showing a progressive fall in extrathyroidal store of thyroxine overtime.

Thyroxine acts on almost all organs of the body including the gut and regulates metabolism. The results provide detailed information of the activity of different parts of the intestine for glucose transfer using the everted sac model. They include nine of the twelve different variables of absorption recommended by Beryl. Although these variables are dependent, it was suggested that data from these variables should form an indispensable baseline for studying the transfer capacity of the intestine to avoid fallacious deductions[6]. The everted sac technique has the advantage that all regulatory factors whose influence may be unwanted or difficult to account for in an invivo study have been eliminated by the isolation of the intestine; it also enables determination of regional variation in absorption in the different part of the intestine[7].

The changes in glucose concentration in mucosal and serosal fluids depended on the relative movements of fluid and glucose. In all the sacs FMC was lower than the initial concentration showing a positive glucose transfer from the mucosal fluid. However, not all these glucose appear in the serosal fluid. Much more glucose disappeared from the mucosal fluid than appeared in the serosal fluid. This may be due to the metabolism in the intestine of part of the glucose disappearing from the mucosal fluid. Hyperthyroid group had the greatest percentage MGT while hypothyroid group had the least.

There was a slight reduction in the serosal glucose concentration. However, FSC was greater than FMC giving a net positive FCG and glucose transport. FCG in the hypothyroid was reduced compared to the control in a manner similar to that reported for phlorizin inhibition of glucose transport.[8] The reduced gradient results in a decrease in glucose transport across the hypothyroid intestine thus hypothyroidism is characterized by impaired glucose absorption from the gastrointestinal tract. However, FCG was elevated when thyroxine replacement was given to the Euthyroid group showing that thyroxine increases the gradient at which glucose is transported and consequently glucose transport across the intestinal wall. Hyperthyroid intestine had the greatest FCG and thus the greatest glucose transport. This is because the greater the FCG the greater the glucose transport since a steep concentration gradient is necessary for glucose to move appreciably across the submucosal tissues.

FCG and MGT were greater in the jejunum than in the ileum across all the groups giving rise to greater glucose transport in the jejunum than the ileum in all the groups. The increased transport activity in the jejunum than the ileum in all the groups mean that T4 facilitated intestinal glucose transport in a manner that maintained the variational differences in the transfer capacity of the intestine contrary to the suggestion of Spencer *et al*.[9]

Gut glucose uptake (GGU) was significantly low in the hypothyroid group, high in the hyperthyroid group and in the euthyroid group GGU was elevated similar to that of the control. This also may be attributed to the reduced metabolic and thus transfer capacity in hypothyroidism and increased metabolism and transfer capacity by thyroixine. Negative serosal glucose transfer was observed in the hyperthyroid jejunum and ileum and euthyroid ileum. This significant glucose loss from the serosal fluid cannot be solely attributed to the increased metabolism of the enterocytes caused by hyperthyroidism. Excess thyroxine may cause the movement of glucose in the opposite direction (negative SGT) in both the jejunum and ileum while thyroxine replacement may influence the transport of glucose in the reverse direction in the ileum. This means that hyperthyroidism may cause the reverse absorption of blood glucose into the intestine from the serosal side.

CONCLUSION

Thyroxine replacement caused a rise of GGU, FGT, MGT and SGT in a manner similar to control. Hyperthyroid intestine had a greatly increased GGU, FGT, MGT and a negative SGT while in hypothyroid intestine GGU, FGT, MGT and SGT was greatly reduced. MGT, GGU, FGT and SGT was greater in the jejunum than the ileum across all the groups showing a variation in glucose transfer capacity in the different parts of the intestine independent of and potentiated by thyroxine. Thus glucose transfer is greater in the jejunum than ileum in different thyroid states.

Disappearance from the intestinal lumen in the whole animal corresponds to mucosal transfer in vitro, and entry into the blood stream is analogous to serosal transfer.[10] Therefore thyroidectomy causes reduced glucose absorption from the small intestine, thyroxine replacement increases absorption of glucose while excess thyroxine may cause reverse absorption of glucose back into the intestinal enterocytes.

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Electrocardiographic Changes in Normal Pregnancy

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ABSTRACT

Background: Pregnancy is a physiological condition which affects all the functions of maternal body, that brings a considerable alteration in hemodynamic activity of the maternal heart. It is very important to understand the cardiac functioning of maternal heart during normal pregnancy because apart from obstetric causes, cardiac disorders are the main cause of mortality in females. Pregnancy also brings about various changes in the electrocardiogram, further confusing with that of heart disease. This study is undertaken to highlight the effect of normal pregnancy on the Electrocardiogram and thereby helps us to distinguish it from that of pathological changes.

Objectives: To study the effect of normal pregnancy on the electrocardiogram and to compare with that of normal non pregnant women.

Method: 50 normal pregnant women in 2nd and 3rd trimester each between 20- 35 years of age and 50 normal non pregnant women of the same age group were selected for the study. A 12 lead ECG was recorded by using ECG machine with special emphasis on changes in heart rate, PR interval, QT interval, QTC interval, ,and all the parameters were analyzed.

Results: The ECG changes observed in our study include ,significant increase in heart rate and QTC interval, increase in QT interval, significant decrease in PR interval ($p < 0.05$) when compared to normal non pregnant women.

Conclusion: There is alteration in circulatory dynamics during pregnancy which leads to significant variations in ECG from the average normal. Our findings highlight the need for caution in the interpretation of ECG in the antenatal period which in turn help us in better management of those with cardiac disease.

Keywords: *Pregnancy, Heart Rate, PR interval, QTC, Electrocardiogram*

INTRODUCTION

Pregnancy although, a physiological phenomenon affects all the functions of the maternal body,[1]especially changes in the cardiovascular system do occur during normal pregnancy[2] to meet the increased metabolic needs of the mother enabling adequate delivery of oxygenated blood to the peripheral tissues and to the fetus.[3]

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Pregnancy is associated with major hemodynamic and cardiac changes, which can mimic or precipitate cardiac diseases. Pregnancy also brings about various changes in ECG. Probably, the most common reason for referral of the pregnant patient from the obstetrician to the cardiologist is evaluation of a systolic murmur heard over the precordium.[4] To have diagnostic specificity of cardiovascular disease the ECG changes must exceed normal variations encountered during pregnancy. It is important not to diagnose heart disease when none exists and at the same time not to fail to detect and appropriately treat heart disease when it does exist.[5]

The Electrocardiogram (ECG) is a graphical record of the electrical potential caused by the excitation of the cardiac muscle has been shown to be affected by

several physiologic factors like race, age, sex, height, weight, nutritional status, chest circumference & other factors. [6]

The present study was set out to determine electrocardiographic changes in normal pregnancy.

MATERIALS AND METHOD

50 apparently healthy pregnant women in 2nd and 3rd trimester each, between 20- 35 years were selected consecutively as and when they presented to the obstetric outpatient department of Bapuji Hospital and Chigateri Hospital, Davangere. 50 healthy non pregnant women of the same age group were selected randomly from general population. The study was conducted in Physiology department, J.J.M Medical College, Davangere

Participants were given an explanation of the study protocol and informed consent was taken from each subject.

A pretested structured proforma was used to record the relevant information from each individual case selected. Data acquisition was performed in the morning.

A detailed physical and systemic examination was done in subjects who were selected. Physical examination of all the subjects included measuring height in centimeters, weight in kilograms. Subjects were matched for age, height, weight and nutritional status.Information's like the name, age, parity, and gestation were recorded. Subjects were allowed to take rest for ten minutes. Recording of resting pulse rate done by palpating the radial artery and blood pressure was recorded with a mercury sphygmomanometer using the appropriate sized cuff.

Subjects were screened for the presence of inclusion and exclusion criteria and dropped if any exclusion criteria that are likely to affect cardiovascular system were present. Women aged less than 20 and more than 35 years, women with any organic cardiac disease. Renal disease, Severe Anemia, Thyroid disease, Diabetes, Hypertension, with chronic medication and with history of surgery was excluded from this study.Only singleton pregnancies were eligible.

ELECTROCARDIOGRAPHIC RECORDING

A 12 lead electrocardiogram was recorded in the subjects during the resting state and analyzed carefully.

The instrument used to record electrocardiogram is the twelve channel electrocardiograph HEWLETT PACKARD page writer manufactured by Philips Electronics Ltd.

All the measurements were made with the help of magnifying lens in all the leads in which clear curve was obtained.

In the Electrocardiogram following findings are regarded as normal

- Heart rate between 60 to100 bpm.
- P-R interval from 0.12 sec to 0.20 sec.
- QT_C interval: upper limit of about 0.46sec

The heart rate was calculated from the table 3:1 in principles of electrocardiography by Goldman MJ .[7]

QT_C interval is determined using modified Bazett's formula by Hodges and co-workers. It corrects more completely for high and low rates. [8]

Statistical Analysis

Results were expressed as Mean ± SD for continuous data, number and percentages for categorical data. One way ANOVA was used for multiple group comparisons followed by 'Post – hoc – Tukey' test for group – wise comparisons. Categorical data was analyzed by Chi – square test. A 'p' value of 0.05 or less was considered for statistical significance i.e.,

$p < 0.05$, $p < 0.01$: S – Significant.

$p < 0.001$: HS – Highly Significant.

$p > 0.05$: NS – Not **Significant.**

RESULTS

The statistical comparisons of the matching variables (age, height and weight) are inherently similar for all groups. The following results were obtained in our study.

Table 1 shows comparison of heart rate expressed in bpm among the three groups i.e. . Pregnant women in 2nd trimester, pregnant women in 3rd trimester and the normal non pregnant women.

Heart rate showed significant increase in both the pregnant groups when compared to controls ($p < 0.01$). There was also significant increase in heart rate in

pregnant women in third trimester when compared to pregnant women in second trimester (p<0.01).

Table 2 represents comparison of PR interval (seconds) among three groups.

PR interval showed significant decrease in both pregnant women group when compared to controls (p < 0.01).There was also significant decrease in PR interval in pregnant women in 3rd trimester when compared to pregnant women in 2nd trimester (p <0.01).

Table 3 shows comparison of QT interval (sec) among the three groups.

In this study though there was slight increase in QT interval in pregnant women in third trimester, there was no statistical significance when values of controls, pregnant women in 2nd trimester and pregnant women in 3rd trimester were compared (p > 0.05).

Table 4 shows comparison of QT_C interval values (sec) among the three groups.

In this study QT_C showed significant increase in pregnant women in both trimester when compared to controls (p <0.01). There was also significant increase in QT_C interval in pregnant women in 3rd trimester when compared to pregnant women in 2nd trimester (p < 0.01).

In spite of these findings, there was no difference in the overall impression regarding the normality of the ECG between either of the two stages of pregnancy and controls.

Table 1: Comparison of heart rate (bpm) between three groups

Variables	Particulars	Controls[A]	2nd TM[B]	3rd TM[C]	Difference between groups			
					F value	A-B	A-C	B-C
HR (in bpm)	Mean ± SD	79.0 ± 6.60	87.2± 6.16	94.2 ± 8.5	F - 56.06p <0.05HS	p < 0.01 S	p < 0.01 S	p< 0.05 S
	Range	68-100	76-104	68-110				

Table 2: Comparison of pr interval (sec) between three groups

Variables	Particulars	Controls[A]	2nd TM[B]	3rd TM[C]	Difference between groups			
					F value	A-B	A-C	B-C
PR Interval (in sec)	Mean ± SD	0.15 ± 0.01	0.13± 0.02	0.11± 0.02	F - 19.8p <0.05HS	p < 0.01 S	p < 0.01 S	p< 0.05 S
	Range	0.12 – 0.16	0.10 - 0.16	0.10 - 0.16				

Table 3: Comparison of QT interval (sec) between three groups

Variables	Particulars	Controls[A]	2nd TM[B]	3rd TM[C]	Difference between groups			
					F value	A-B	A-C	B-C
QT Interval (in sec)	Mean ± SD	0.35± 0.02	0.35 ± 0.01	0.36 ± 0.01	F- 0.72p > 0.05NS	NS	NS	NS
	Range	0.32 - 0.36	0.32 - 0.36	0.32 - 0.36				

Table 4: Comparison of QTc interval (sec) between three groups

Variables	Particulars	Controls[A]	2nd TM[B]	3rd TM[C]	Difference between groups			
					F value	A-B	A-C	B-C
QT_C Interval (in sec)	Mean ± SD	0.38 ± 0.01	0.40 ± 0.01	0.43± 0.01	F - 61.44p <0.001HS	p < 0.01 S	p < 0.01 S	p< 0.05 S
	Range	0.35 -0.41	0.37 - 0.42	0.38 - 0.44				

DISCUSSION

This study demonstrated that certain ECG measures are altered by the physiological state of pregnancy in the absence of demonstrable heart disease. To the best of our knowledge the reasons for electrocardiographic changes during pregnancy may be due to:

1. The changed spatial arrangement of the chest organs.
2. Changed electrical properties of the myocardium due to changes in both the sympathetic and hormonal modulation (epinephrine, progesterone) of the electrical heart activity during pregnancy.

Further we noticed a significant change in electrical activity in pregnancy in terms of heart rate PR, QT and QT_C intervals. These changes became more significant with progression of pregnancy and peaked during the 3rd trimester.

Interestingly, pregnancy may be associated with a concentric enlargement of the left ventricle in response to the hemodynamic requirements, which in turn could explain this ECG changes.[9]

The effect of normal pregnancy on electrocardiogram is analyzed as follows.

Heart rate

In our study there was significant increase in heart rate with progression of pregnancy compared to normal non pregnant women.

Hunter S and his colleagues in their study found that, the heart rate increase was seen by 5th week of gestation and continued till 32 Weeks.[10]

The heart rate increases gradually throughout pregnancy, reaching a level that is approximately 25% above the non pregnant levels at the time of delivary.[11]

Furthermore Charles B& his co-workers in their study found that among the pregnant women studied, at six and half months of gestation the mean cardiac rate was 99 per minute; by 8 months of gestation the mean cardiac rate had increased to 109 per minute in their study, but in early labor it slowed to 95 per minute.[12]

While considering the above findings, increase in heart rate in pregnancy can be linked to autonomic nervous system changes that produce alterations in cardiac autonomic modulation. [13]

The increase in heart rate in early pregnancy is linked to hormonal mechanism. In relation to endocrinological factors in early pregnancy, the initial change in heart rate may be linked to the production of chorionic gonadotropin, with the later gradual increase being related to the vascular changes which accompany placental and fetal growth.[14]

In addition, this increase in heart rate mainly during third trimester compensates for the fall in stroke volume resulting from caval compression.[15] Also the cardiac output increases as early as 5 weeks and rises to 45% above the baseline at 24 weeks of gestation. This is achieved by increase in heart rate and stroke volume.[16]

PR interval

In the current study it was observed that, PR interval showed significant decrease in pregnant women in both the trimesters when compared to controls. There was significant decrease in PR interval in 3rd trimester compared to 2nd trimester.

Study by Carrutn JE and his colleagues found that the mean PR interval was shorter at third trimester compared to first and second trimester..[6]

The decrease in PR interval during pregnancy could be due to shortening of A-V conductance with respect to increase in heart rate that accompany during pregnancy. [17]

QT interval

In this study there was slight increase in QT interval in pregnant women in 2nd trimester and 3rd trimester when compared to controls but the change was not found to be significant.

QT_C Interval

QT_C Interval in the electrocardiogram reflects the time taken for depolarization and repolarization in the ventricular myocardium. The QT Interval when corrected for heart rate is (QT_C).

In the current study, though QT interval (Sec) did not show any significant increase in pregnancy, it was found that when QT interval was corrected with heart rate, which was increased to a large extent in our study as, pregnancy progressed.

In control group, the QTc is about 0.39 sec but in pregnant group QTc gradually increased to about 0.41 to 0.43 sec. There is increase of about 0.04 sec. According to Goldman M.J. et. al [10] average QTc interval in non-pregnant female is about 0.394 sec.

Study by Lechmanova M et al reported increased heart rate, shortening of A-V conductance, prolongation of QT interval normalized for heart rate, changes in the ventricular depolarization and repolarization patterns were observed. [18]

Lechmanova M & Kittnar O and their co-workers in their study evaluated the possible physiological

determinants of QT prolongation. There was an increase in QT interval as well as prolongation of QTc interval during late pregnancy. The possible causes prolongation of QT and QTc interval was attributed to changed spatial arrangement of chest organs during pregnancy, changed electrical properties of myocardium due to changed sympathetic and hormonal modulation of the electrical heart activity, alteration in circulatory dynamics which leads to significant variation in duration of electrical systole from average normal.[19]

However, the differences observed between the mean ECG measures of the pregnant population and those of the normal population are not clinically significant.

Previous studies have suggested that, sex hormones (estrogen, progesterone) and serum electrolyte changes during pregnancy can affect the ECG. This study gives quantitatively and qualitatively more effective results, if hormonal assay as well as serum electrolytes assessment is included along with the study. and also if the same subjects were taken as controls before pregnancy and were followed during pregnancy. Hence further studies are needed by considering these facts to evaluate the effect of normal pregnancy on electrocardiogram and to explore the supposed mechanisms.

CONCLUSION

In conclusion we report, ECG changes like significant increase in heart rate, decrease in PR interval, increase in QT & QT_C interval in pregnant women compared to non pregnant women.

This study concludes that there is a need for systematic evaluation of hemodynamic changes and also need for caution in the interpretation of ECG during antenatal period in view of the considerable variability of the electrocardiogram during normal pregnancy.

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Effect of Visfatin on Blood Pressure in Normal and DOCA-Salt Hypertensive Rats

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ABSTRACT

Background and aim: Visfatin is an adipokine secreted by adipose tissue. Visfatin has been reported to be involved in several biological processes in the cardiovascular system. However, the effect of visfatin on blood pressure is still unclear. In this study, we examined the effect of visfatin on DOCA-salt hypertensive rats and normotensive control rats.

Method: 48 rats were randomized into eight groups (n=6, each), 1- control group. 2- Visfatin treated group 3- Uninephrectomized(UNX) group. 4- UNX plus visfatin treated group. 5- DOCA-salt group. 6- DOCA-salt plus visfatin (1nM) treated group 7- DOCA-salt plus visfatin (10nM) treated group and 8- DOCA-salt plus visfatin (100nM) treated group. Systolic and diastolic blood pressures were recorded every week during the entire period of the study by tail-cuff method.

Results: Visfatin (1nM) produced insignificant inhibitory effect on blood pressure; on the other hand, visfatin (10nM) produced significant and (100nM) highly significant inhibitory effect on systolic and diastolic blood pressures in DOCA- salt hypertensive rats.

Conclusion: The results of the present study demonstrated that visfatin has significant inhibitory effect on the elevated blood pressure in DOCA-salt hypertensive rats. Further studies needed to clarify the detailed mechanisms by which visfatin decreases the blood pressure.

DOCA: DeOxyCorticosteroneAcetate.

Keywords: *Visfatin - Hypertension- hypertensive Rats- Metabolic Syndrome*

INTRODUCTION

Visfatin (also known as nicotinamide phosphoribosyltransferase and pre-B cell colony-enhancing factor) is an adipokine secreted mainly by visceral adipose tissue. Its binding to the insulin receptor induces insulin like action [(1)]. Numerous studies have been published to address the possible associations between plasma visfatin levels and various metabolic disorders such as obesity, type 2 diabetes [(2, 3)], and metabolic syndrome [(4)].

In the cardiovascular system, it has been reported that visfatin produces cardioprotection in ischemic myocardial injury [(5)], induces vascular endothelial angiogenesis [(6)]. Mazaherioun et al., detected high levels of visfatin in patients with acute myocardial infarction and concluded that proinflammatory cytokines such as visfatin may play a role in atherosclerosis as well as destabilization of the atherosclerotic plaque [(7)]. It was also demonstrated that perivascular adipose visfatin regulates vascular smooth muscle cell growth and apoptosis via both endocrine and paracrine pathways [(8)].

Little is known about the effect of visfatin on blood pressure in normal and induced hypertensive rats. In a small sample in clinical research, Dogru et al. [(9)] have shown that there was no change of blood visfatin levels

in a specifically selected group of young hypertensive patients without obesity or lipid and glucose metabolic abnormalities. Other study reported that serum visfatin was increased and associated with lipid metabolic abnormality in Lyon hypertensive (LH) rats [10]. In another study [11] it was found that no significant changes of circulating visfatin and no association between circulating visfatin levels and blood pressure in normotensive and spontaneously hypertensive rats. However, Gunes et al. [12] found that the mean visfatin level was significantly higher in hypertensive patients and the serum visfatin levels in the pre-hypertensive group were also significantly higher than in participants with normal blood pressure. They also found a significant positive correlation between visfatin and blood pressure [12]. On the other hand other study [13] reported that increased plasma visfatin concentration may play a significant role in the pathogenesis of hypertension in patients with visceral obesity.

For all the above mentioned controversy about the role of visfatin in blood pressure regulation and because the studies made on the effect of visfatin on blood pressure were deficient, we designed this study to elucidate the effect of visfatin on blood pressure in normotensive and hypertensive rats.

MATERIALS AND METHOD

Animals: 48 male albino rats (11-13 weeks old, 180–220 g) were included in the experiment obtained from the Laboratory Animal Research Unit of Zagazig University, Egypt. Rats were housed in a 12:12-h light-dark cycle at 25 ± 2°C, and had free access to tap water and standard rat chow *ad libitum* for 7 days to acclimatize before entering the study. All experiments were performed in accordance with the Institutional guidelines for the Care and Use of Animals for Scientific Purposes and in accordance with the recommendations from Helsinki Declaration.

Drugs and chemicals: visfatin and DOCA were purchased from (Sigma Chemical Co., St. Louis, MO, U.S.A.). Visfatin was dissolved in normal saline and prepared freshly at the desired concentrations [5], while DOCA was dissolved in DiMethyle SulfOxide (DMSO)[14].

Method of uninephrectomy

Left uninephrectomy was performed on 36 rats. Rats were anaesthetized with intraperitonial injection of ketamine (75 mg/kg), kidney was visualized by a left lateral abdominal incision (1 cm long), and the left renal artery and ureter were ligated by silk thread, and then the left kidney was removed and weighed. The muscle and skin layer (incision site) were sutured with highly sterile suture needles [14].

DOCA-salt hypertensive rats:

After uninephrectomy, UNX-rats were allowed to drink tap water *ad libitum*, with no further treatment. In DOCA-salt hypertensive groups, rats were given 1% NaCl in the drinking water with subcutaneous injection of DOCA (25 mg/kg, twice weekly) with mild heating for six consecutive weeks [15].

Experimental protocol

1. **Control group:** given normal saline.
2. **Visfatin treated group:** given 100 nM visfatin.
3. **Uninephrectomized (UNX) group:** given vehicle.
4. **UNX plus visfatin treated group:** the rats were injected intravenously with visfatin (100 nM) once each morning (9-10 a.m.) [16].
5. **DOCA-salt hypertensive group:** given subcutaneous injection of DOCA (25 mg/kg, twice weekly).
6. **DOCA-salt plus visfatin (1nM) treated group:** given visfatin 1nM [1].
7. **DOCA-salt plus visfatin (10nM) treated group:** given visfatin 10nM [16].
8. **DOCA-salt plus visfatin (100nM) treated group:** given visfatin 100nM.

Measurement of blood pressure: systolic and diastolic blood pressures were recorded every week during the entire period of the study by tail-cuff method (IITC, model 31, Woodland Hills, CA, USA). The animals were placed in heated chamber at an ambient temperature of (30–34 °C) for 15 min and from each animal; 1–9 blood pressure values were recorded. The lowest three readings were averaged to obtain a mean blood pressure. All the recordings and data analyses were done using a computerized data acquisition system and software [17].

Statistical analysis: All data were presented as mean ± standard deviation. Statistical significance of

the differences was calculated by unpaired Student's *t*-test using SPSS version 19 (SPSS for Windows). A *p* value d" 0.05 was considered statistically significant.

RESULTS:

Table 1 and Table 2: show that the systolic and diastolic blood pressures were significantly increased in DOCA-salt hypertensive rats compared to UNX-rats ($P<0.05$). Visfatin (1 nM) produced insignificant inhibitory effect on both systolic and diastolic pressures in DOCA salt hypertensive group compared to the control. Visfatin (10 nM) for a period of six consecutive weeks significantly decreased systolic and diastolic blood pressure in DOCA-salt treated rats ($P<0.05$). Visfatin (100 nM) produced highly significant inhibitory effect on systolic and diastolic blood pressure in DOCA-salt treated rats ($P<0.005$).

Table 1: effect of visfatin on systolic blood pressure (mmHg):

	DOCA	DOCA+V1	DOCA+V10	DOCA+V100
Day1	121±4	120±6	119±3	118±3
1st w	140±6	135±4	132±4*	125±5**
2ndw	150±7	142±6	141±4*	130±3**
3rd w	160±8	158±8	150±7*	125±4**
4th w	170±9	167±6	158±7*	130±5**
5th w	180±9	176±8	167±9*	132±5**
6th w	190±8	185±9	180±6*	143±5**

DOCA: Deoxy-corticosterone acetate

V1: visfatin 1nM. V10: visfatin 10nM. V100: visfatin 100nM.

* = significant (P < 0.05) compared to DOCA group ** = highly significant (P< 0.01) compared to DOCA group

Table 2: Effect of visfatin on diastolic blood pressure (mmHg)

	DOCA	DOCA+V1	DOCA+V10	DOCA+V100
Day1	85±4	85±4	84±3	83±3
1st w	100±7	95±7	90±6*	85±6**
2ndw	115±6	110±4	107±4*	92±3**
3rd w	120±6	115±5	113±4*	94±4**
4th w	130±7	120±6	115±5*	97±5**
5th w	140±9	135±8	125±6*	100±6**
6th w	145±8	140±9	134±7*	105±6**

DOCA: Deoxy-corticosterone acetate

V1: visfatin 1nM. V10: visfatin 10nM. V100: visfatin 100nM.

* = significant (P < 0.05) compared to DOCA group ** = highly significant (P< 0.01) compared to DOCA group

Table 3 and 4: show that Visfatin produced insignificant inhibitory effect on systolic (table and figure 3) and diastolic (table and figure 4) blood pressures in normal and uninephrectomized (UNX) rats.

Table 3: effect of visfatin on systolic blood pressure (mmHg)

	N	N+V100	UNX	UNX+V100
Day1	110±2	108±2	120±4	118±5
1st w	105±1	104±1	121±5	119±4
2ndw	112±3	110±3	122±3	120±4
3rd w	115±4	111±4	120±4	118±4
4th w	110±3	108±3	122±5	121±5
5th w	120±5	116±5	121±6	119±4
6th w	118±6	115±6	122±5	120±5

N: Normal. V100: visfatin 100 nM.

UNX: uninephrectomized.

Table 4: Effect of visfatin on diastolic blood pressure (mmHg)

	N	N+V100	UNX	UNX+ V100
Day1	78±1	77±1	80±2	78±3
1st w	80±2	78±2	82±2	81±3
2ndw	75±2	74±3	81±2	80±4
3rd w	80±3	78±3	82±1	81±4
4th w	81±3	80±4	82±2	80±3
5th w	82±4	81±5	83±3	81±4
6th w	80±3	78±4	83±4	82±5

N: Normal. V100: visfatin 100 nM.

UNX: uninephrectomized.

DISCUSSION

Visfatin is an adipokine secreted mainly by visceral adipose tissue. In the cardiovascular system, it has been reported that visfatin produces cardioprotection in ischemic myocardial injury [5], exhibited direct cardioprotective effects [18]. Many ctions of visfatin have been linked to cardioprotection [19,20]. However, the role of visfatin in regulating blood pressure is still in controversy, so, we focused in this study on the effect of visfatin on systolic and diastolic blood pressure in normotensive and DOCA salt induced hypertensive rats.

In our study, we found that visfatin has insignificant inhibitory effect on blood pressure in normal rats. We found also that visfatin (1 nM) produced insignificant inhibitory effect on systolic and diastolic pressures in DOCA salt treated rats. Our results in agreement with Wang et al., [11] who found that no significant changes of circulating visfatin and no association between circulating visfatin levels and blood pressure in normotensive and spontaneously hypertensive rats. However, we found significant ($P < 0.05$) inhibitory effect of visfatin (10 nM) on both systolic and diastolic pressures in DOCA salt induced hypertensive rats. These findings were in controversy with the findings of Wang et al., [11], but we can explain this controversy by the different animal model, where they used spontaneously hypertensive rats instead of DOCA salt hypertensive rats in our study.

In addition, we found that visfatin (100 nM) produced highly significant ($P < 0.005$) inhibitory effect on systolic and diastolic pressures in DOCA salt treated rats. Our results in agreement with Gunes et al., [12] who found that the mean visfatin level was significantly higher in hypertensive patients and the serum visfatin levels in the pre hypertensive group were also significantly higher than in participants with normal blood pressure. They also found a significant positive correlation between visfatin and blood pressure.

We can explain the elevated serum levels of visfatin in hypertensive patients by the suggestion that visfatin act to counteract the elevated blood pressure and as a compensatory mechanism done by visceral adipose tissue to decrease the high blood pressure to its normal value. Our suggestion is supported by the findings of Mazaherioun et al., [7] who detected high levels of visfatin in patients with acute myocardial infarction. They concluded that pro-inflammatory cytokines such as visfatin may play a role in the development of atherosclerosis as well as destabilization of the atherosclerotic plaque.

Also, our results were found to be in agreement with Filippatos et al., [4] who found that plasma visfatin levels are increased in patients with metabolic syndrome compared with individuals that do not fulfill the criteria for this syndrome. However our results were in controversy with Kloting and Kloting [21] who found that visfatin gene expression in visceral and subcutaneous adipose tissue is similar to that in lean control animals in the Wistar Ottawa Karlsburg rat, a model of polygenic metabolic syndrome with obesity, hypertension, dyslipidemia, hyperinsulinemia, and impaired glucose tolerance. However this controversy can be explained by the differences in used animals where we used normal male albino rats where they used a rat model with polygenic metabolic syndrome.

Also, our results were in agreement with Rotkegel et al., [13] who found that hypertensive patients had significantly higher plasma visfatin level than the control group. However, they suggested that increased plasma visfatin concentration may play a significant role in the pathogenesis of hypertension in patients with visceral obesity. In controversy, we suggest that

increased plasma visfatin level is a result not a cause and we assumed that visfatin secretion is increased to decrease the elevated blood pressure. Our last suggestion is supported by Yamawaki et al., [22](who found that pretreatment with visfatin (100 ng/ml, 30 min) inhibited noradrenaline (NA; 1 nM-1 microM)-induced contraction in endothelium-intact rat aorta. They also found that visfatin (1-100 ng/ml) directly induced a relaxation in NA (100 nM)-pre-contracted aorta.

In conclusion, this study proved that visfatin has an inhibitory effect on systolic and diastolic blood pressures in DOCA salt hypertensive rats in a dose dependent manner. We suggested that visfatin may have a protective role against hypertension and this role should be further evaluated by human studies.

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Co-Relation Study of Dietary Habits and Early Renal Damage in Hypertensive Patients

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ABSTRACT

Hypertension is a result of complex interactions between non-modifiable genetic and other modifiable risk factors. Dietary habits play a vital role in modifiable risk factors; as non-vegetarian increases the risk of hypertension and results in cardiovascular, renal and other manifestations. In our study a total of 60 hypertensive patients of both sex were selected randomly of whom 30 were on vegetarian diet (group I) and 30 were on mixed diet (group II). The high sensitive C-reactive protein (hsCRP) and urine microprotein levels were more increased in group II than group I hypertensive patients and was statistically significant. When gender-wise comparision of hsCRP and urine microprotein levels in group I and group II hypertensive patients was done, it was more increased in group II hypertensive patients than group I and was not statistically significant Hence the present study helps to understand that, a simple change from mixed diet to vegetarian diet is helpful to control the extent of renal damage and other associated complications in hypertensive patients.

Keywords: *Hypertension, High sensitive C reactive protein, Microprotein*

INTRODUCTION

Hypertension is one of the most common non-communicable diseases with high morbidity and mortality rate.[1] Hypertension is a result of complex interactions between non-modifiable (genetic, age, gender) and other modifiable risk factors. Dietary habits play a vital role in modifiable risk factors; as non-vegetarian diet increases the risk of hypertension and results in cardiovascular, renal and other manifestations.[2] Microproteinuria has been shown to be a reliable index to predict the development of early renal damage.[3] Also inflammatory markers like C-reactive protein (CRP) are also found to be raised in hypertension.[4] Hence the present study was undertaken to study the relation between dietary habits and CRP levels and also to compare the degree of early renal damage in hypertensive patients on vegetarian and mixed dietary habits.

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OBJECTIVES

To determine whether there is any relationship between hypertension and C-reactive protein levels, hypertension and renal damage and also to compare the same in patients on vegetarian and mixed dietary habits so as to emphasise the role of change in dietary habits in controlling the complications of hypertension.

METHODOLOGY

The present study was conducted in a tertiary referral hospital in hypertensive patients in the age group of 35 to 60 years of both gender. A total of 60 patients diagnosed as hypertensive were selected randomly of whom 30 were on vegetarian diet (group I) and 30 were on mixed diet (group II) using inclusion and exclusion criteria.

Each subject gave an informed written consent and the protocol of the study was explained in detail in vernacular language. The study was approved by the Institutional Ethical Committee.

Inclusion Criteria includes patients with h/o hypertension for more than 2 years, pure vegetarian diet (group I) and on mixed diet (patients on non-vegetarian meals for atleast 2 times/ week since 10 years) (group II). Exclusion criteria includes H/O diabetes mellitus, alcohol intake, smoking, uncontrolled hypertension, on irregular antihypertensive treatment, fever in the past week and other medical illness.

A detailed medical and dietary history was obtained and detailed medical examination was done. Two millitre of blood sample was collected under aseptic precautions, allowed to clot for 20 minute, centrifuged for 10 minute at the rate of 3000 rpm to separate the serum which was used to assess CRP levels by using semi-autoanalyser. About 5 ml of midstream urine sample was collected under hygienic conditions in a clean container for estimation of microproteinuria. The statistical analysis was done by using Student 't' test was for comparing two groups.

RESULTS AND DISCUSSION

Hypertension is a chronic medical condition of the cardiovascular system. It is one of the most common non-communicable disease with high mortality and morbidity rates.[5] Hypertension solely is responsible for 12.8% of global deaths in 2004, 51% of it from stroke and 45% of it from coronary heart disease.[6] A study conducted by ICMR during 2007-2008 revealed 17-21% of prevalence of hypertension in all the states of India.[7]

Hypertension is influenced by various risk factors. The non-modifiable risk factors are age, sex and genetic factors and the modifiable risk factors are diet, alcohol, smoking, environmental factors etc.[8]

Diet is one of the very important modifiable risk factor as non-vegetarian diet yields more saturated fats that increases the plasma cholesterol level and hence predisposes the individual for the development of hypertension.[9]

Table 1. Shows the comparison of hsCRP and urine microprotein levels in group I and group II hypertensive patients

	Group I	Group II
Mean hsCRP (mg/dL)	3.46 ± 0.96	4.26±1.71 *
Mean urine microprotein (mg/dL)	29.97 ± 15.32	41.48±20.82*

* P<0.05 – statistically significant

Table 2. Shows the gender-wise comparison of hsCRP levels in group I and group II hypertensive patients

Gender	Group I		Group II	
	Number of Patients (n=30)	Mean hsCRP (mg/dL)	Number of patients (n=30)	Mean hsCRP (mg/dL)
Males	18	3.40	21	4.69
Females	12	3.27	09	3.55

P>0.05 – statistically not significant

In the present study the acute phase inflammatory marker, high sensitive C-reactive protein (hsCRP) was more increased in group II hypertensive patients than group I and was statistically significant (table 1) and is in association with other studies.[10,11,12]

When gender-wise comparison of hsCRP levels in group I and group II hypertensive patients was done, it was more increased in group II hypertensive patients than group I and was not statistically significant (table 2)

C-reactive protein is a protein synthesized in the liver whose levels are increased in the blood in response to inflammation.[10] Studies have also been conducted to know its direct relation with prediction of hypertension.[11] Studies also reveal that C-reactive protein can itself be an independent risk factor in the development of hypertension.[13]

Hypertension per se may leads to multiple inflammatory stimuli at the vessel wall which in turn promote the production of a number of pro inflammatory cytokines such as tumor necrosis factor, interleukin-6 and CRP as a defense against injurious factors. Inflammation, common in hypertensives, decreases endothelium dependent relaxation, possibly by decreased capacity of the endothelium to generate vasodilatory factors like nitric oxide (NO) which inturn raises blood pressure. [13]

Table 3. Shows gender-wise comparison of urine microprotein levels in group I and group II hypertensive patients

Gender	Group I		Group II	
	Number of Patients (n=30)	Mean urine microprotein (mg/dL)	Number of patients (n=30)	Mean urine microprotein (mg/dL)
Males	18	34.906	21	45.800
Females	12	22.586	09	30.680

P>0.05 – statistically not significant

In the present study the reliable marker of kidney damage, microprotein was more excreted in the urine of group II hypertensive patients than group I and was statistically significant (table 3) and is in accordance with other studies.[14,15]

When gender-wise comparison of urine microprotein levels in group I and group II hypertensive patients was done, it was more increased in group II hypertensive patients than group I and was not statistically significant (table 1)

Persistent elevation of hypertension leads to development of various complications affecting most of the organ systems of the body. There is high risk of developing renal damage if hypertension persists for a long duration which is termed as hypertensive nephropathy. If unchecked, hypertensive nephropathy can result in end stage renal disease.[3, 16]

Scope for further study: Hypertension is a silent killer as it is often asymptomatic. Hence there is a need for expanding the knowledge to know various markers associated with it. Hypertension is not a curable disease but its complications can be delayed by modification of various modifiable risk factors like dietary habits. Hence there is need for better understanding of the relationship between hypertension and the dietary habits. This made the study important to be conducted.

CONCLUSION

Hypertension is not a curable disease and its prevention is a very difficult task. But delaying its complications can be easily achieved by control over hypertension and simple modification of modifiable risk factors like diet. Hence the present study helps to understand that, a simple change from mixed diet to vegetarian diet is helpful to control the extent of renal damage and other associated complications in hypertensive patients.

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Comparative Assessment of Stroke Volume by Transthoracic Electrical Bio-Impedance & Echocardiography in Patients of Acute Myocardial Infarction

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ABSTRACT

Objectives: To compare non-invasive determination of stroke volume (SV) by whole body electrical bioimpedance using the NICOMON (Larsen & Toubro Ltd., India) apparatus and Doppler echocardiography in acute myocardial infarction (AMI) patients.

Subjects and method: The present study enrolled 100 AMI patients, who have been assessed by ECHO and NICOMON for SV, where ECHO is considered as a reference method for comparison. SV by Transthoracic electrical bio-impedance was measured by passing an alternating current and measuring the bio-impedance across the thorax. End diastolic volume (EDV), End systolic volume (ESV) & Left ventricular outflow tract (LVOT) diameter, measured by ECHO was used to calculate SV. Various statistical methods like "t"-test & correlation coefficient (r) have been used where found suitable.

Results: Mean value of SV by NICOMON was found to be significantly higher as compared to that of ECHO ($p < 0.001$) and a weak positive correlation ("r" value=0.14) between ECHO and NICOMON SV value was found. Conclusion: Measurement of SV by NICOMON showed weak positive correlation with Doppler derived SV. So NICOMON needs more elaborative studies to establish it as an alternative method of ECHO for stroke volume measurement non-invasively.

Keywords: *Transthoracic Electrical Bio-impedance*

INTRODUCTION

Hemodynamic assessment of critically ill patients with cardiac pathology is one of the challenges of recent diagnostics and treatment strategies. Different methods for the assessment of central hemodynamics are used in clinical practice. However, these methods (invasive and non-invasive) have specific advantages and disadvantages.

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Pulmonary artery Swan–Ganz catheters (PAC) are an invasive method for the evaluation of hemodynamics were introduced into clinical practice in 1970[1]. The method of thermodilution, using PAC became a "gold standard method" for the evaluation of hemodynamic changes[2-5].

Sandham JD et al found a significant increase in number of pulmonary artery embolisms in patients using PAC in comparison with the control group[6].

A number of noninvasive methods of assessing hemodynamic parameters have been studied in the past, with transesophageal Doppler echocardiography, impedance cardiography, and carbon dioxide breath analysis currently available. Impedance cardiography

(ICG) is a noninvasive modality that utilizes changes in impedance across the thorax to assess hemodynamic parameters.

Thoracic bio-reactance is a noninvasive method that analyses intrabeat variations of transthoracic voltage in response to an injected high-frequency current. Preclinical and clinical studies have validated the basic principles of this technique and have found a good correlation between bio-reactance and other methods[7-11].

In a multicenter study of noninvasive monitoring systems as alternatives to invasive monitoring of acutely ill emergency patients, Shoemaker WC, et al[12] found that noninvasive monitoring systems gave continuous displays of physiologic data that provided information allowing early recognition of low flow and poor tissue perfusion that were more pronounced in the non-survivors. Noninvasive systems may be acceptable alternatives where invasive monitoring is not available. Invasive hemodynamic monitoring in the ICU provides a series of snapshots at infrequent intervals. Noninvasive monitoring provides similar information for acutely ill patients as continuous, on-line, real time displays anywhere in the hospital. Minor differences between impedance and thermodilution measurements were offset by the advantages of continuous graphic displays of data. They found noninvasive monitoring was easier, quicker, cheaper, and safer than invasive monitoring, but prospective clinical trials are needed to evaluate their cost-effectiveness.

However, in a study H. W. K. NG, et al[13] compared transthoracic bio-impedance and dual beam Doppler ultrasound measurement of cardiac function in healthy volunteers. They concluded that both techniques were able to detect physiological changes in SV and CO in healthy volunteers & the changes detected by both techniques correlate but do not agree. Both methods were suitable to measure individual changes in SV and CO under passive physiological stress but less suitable during exercise.

This study was undertaken to compare the accuracy and precision of measurement of stroke volume by transthoracic electrical bioimpedance (TEB) using a novel instrument (NICOMON, Larsen & Toubro Ltd., India) in comparison with echocardiography method.

MATERIAL AND METHOD

This cross-sectional study was performed after obtaining approval from the institutional review board of KGMU, Lucknow and written, informed consent from the patients. The study was undertaken in 100 patients of acute myocardial infarction, admitted to cardiology emergency of Gandhi Memorial & Associated Hospital and their stroke volume were measured with ECHO and NICOMON. Exclusion criteria were: Patients having cardiac conditions which affect stroke volume like anaemia, valvular heart disease, myocarditis, cardiac tamponade, cardiac metabolic derangements, endocrinal disorders like thyroid dysfunction, arteriovenous fistula (shunt), vitamin deficiency like vitamin B_1 deficiency, pericardial effusion & Patients having conditions which interfere with bio-impedance signal like obesity or pleural effusion.

Stroke volume was measured using the NICOMON instrument (Larsen and Toubro Ltd., India). NICOMON works on the principle of impedance plethysmography. In impedance cardiography four pairs of electrodes measure different haemodynamic parameters. Each pair of electrodes comprises of a transmitting and sensing electrodes. Two pairs are applied to the base of the neck and two pairs are applied at the level of the sternal-xiphoid process junction, directly opposite from each other. The electrodes define the upper and lower limits of the thorax and the distance between them is the thoracic length (L). A high frequency, low amplitude alternating current is introduced through the transmitting thoracic electrodes and the sensing thoracic electrodes measure impedance associated with the pulsatile blood flow in the aorta. By measuring the impedance change generated by the pulsatile flow and the time intervals between the changes, SV can be calculated. The change in impedance is measured from the baseline impedance (Z_0) (overall thoracic resistance to flow of electrical current). It predominantly reflects total thoracic fluid volume. The magnitude and rate of the impedance change is a direct reflection of left ventricular contractility. This change in impedance related to time (dZ/dt) generates a waveform that is similar to the aortic flow curve. The SV can be determined from the impedance curve by extrapolating to the impedance change (dZ) that would result if no blood were to flow out of the thorax during systole. The SV is calculated using the formula,

$$\text{Stroke Volume} = \rho\frac{L^2}{Z_0^2} \times (dZ/dt)_{max} VET$$

where ñ - resistivity of blood, L - mean distance between the inner electrodes (the thoracic length), VET - ventricular ejection time, $(dZ/dt)_{max}$ - the absolute of the maximum value of the first derivative during systole and Z_0 - basal thoracic impedance. VET is obtained from the dZ/dt versus time curve.[14]

Ejection fraction, end diastolic volume, end systolic volume and left ventricular outflow tract diameter determined stroke volume (SV) by Echo method and then SV has been obtained by subtracting ESV from EDV (SV = EDV-ESV).

Statistical analysis of data obtained was done by paired "t"-test to compare the stroke volume by NICOMON and ECHO. The changes in SV were compared using correlation analysis. A "p" value of less than 0.05 was considered to be statistically significant.

FINDINGS

The study was conducted on a cross-sectional design. A total of 100 subjects were enrolled and data from all the 100 patients were used for analysis. The stroke volume and stroke volume index of acute myocardial infarction patients were measured by NICOMON & ECHO and were compared.

Table 1: Comparison of Stroke Volume and Stroke Volume Index between NICOMON and ECHO.

		NICOMON		ECHO		Mean Difference	Statistical Significance		
	n	Mean	SD	Mean	SD		"t"	"r"	"p"
SV (ml/beat)	100	65.5	20.68	48.59	13.33	16.91	6.69	0.14	<0.001
SI (ml/beat/m²)	100	38.58	12.69	28.17	8.84	10.51	6.79	0.24	<0.001

SV mean value by NICOMON was found to be significantly higher as compared to that of ECHO ($p < 0.001$) and a weak positive correlation ("r" value=0.14) between ECHO and NICOMON SV value was found. For the parameter SI mean value in NICOMON was found to be significantly higher as compared to that of ECHO ($p < 0.001$). A weak positive correlation ("r" value=0.24) between ECHO and NICOMON SI value was found.

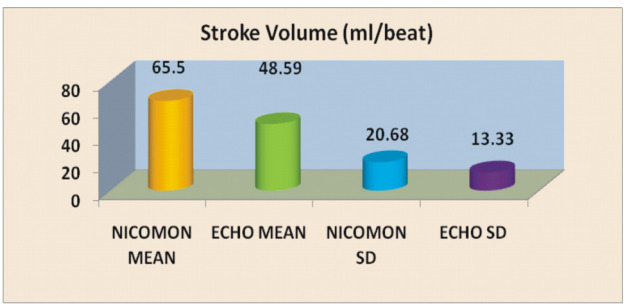

Chart 1: Bar diagram showing comparison of Stroke Volume between NICOMON and ECHO.

Bar diagram showing association between mean values of Stroke Volume obtained by NICOMON (65.5) and ECHO (48.59) with standard deviation of 20.68 & 13.33 respectively.

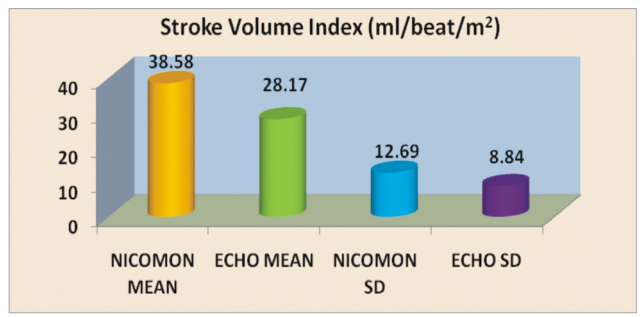

Chart 2: Bar diagram showing comparison of Stroke Volume Index between NICOMON and ECHO.

Bar diagram showing association between mean values of Stroke Volume Index obtained by NICOMON (38.58) and ECHO (28.17) with standard deviation of 12.69 & 8.84 respectively.

DISCUSSION

As the technology advances, there is need for development of minimally invasive & non-invasive techniques for measuring cardiovascular parameters. The thermodilution technique using PAC is invasive, and recently the use for hemodynamic monitoring has been increasingly criticized because of its uncertain risk-benefit ratio and cost[6].

An accurate and reliable noninvasive method for measuring cardiovascular parameters could be a valuable adjunct in the clinical management of patients with acutely decompensated heart failure. Hemodynamic information obtained noninvasively, could help avoid the potentially life-threatening complications of infection, artery perforation, and arrhythmia that can occur with placement of a pulmonary artery catheter. In addition, noninvasive hemodynamic monitoring most likely would decrease costs because of reduced expenditures for equipment.

The results of the present study demonstrate that SV and SI determination by whole-body bio-impedance using the NICOMON device is less accurately correlated with SV and SI determination by Doppler echocardiography ($r = 0.14$ & 0.24 respectively). Also NICOMON SV & SI measurements tended to be significantly higher than Doppler echocardiography readings ($p < 0.001$).

The results of the present study are in concordance with previous study done by Jukka Takala et al[15]. They found that minimally-invasive stroke volume and cardiac output monitoring added to usual care does not facilitate early hemodynamic stabilization in the ICU, nor does it alter the hemodynamic support or outcome. In a study Kazuto Yamashita et al[16] also showed that Transesophageal Doppler echocardiography is a viable minimally invasive method for determining SV and CO in sevoflurane anesthetized beagle dogs, while transthoracic bio-impedance does not provide accurate determination of SV and CO in these dogs

However Talakad N. Sathyaprabha at el[17] in a study found TEB as a good method to determine SV and CO non-invasively in most clinical settings including emergencies. Julija Braždžionyte et al[18] also found in a study that impedance cardiography and transthoracic echocardiography measures of hemodynamic parameters (stroke volume, cardiac output) were significantly correlated.

We can say that data obtained from 100 patients of acute myocardial infarction measured by NICOMON & ECHO shows weak positive correlation and difference between mean values of stroke volume and stroke volume index is significant statistically. So it can be concluded from this study, that NICOMON can be used to measure SV, SI & other cardiovascular parameters, but it does not has advantage over Doppler echocardiography.

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Asymptomatic Postural Orthostatic Tachycardia - A Case Report

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ABSTRACT

We observed orthostatic postural tachycardia in a young adult male aged 19 years. He is completely well and has no symptoms and is carrying out his daily activities comfortably. The data shows that there is marked increase in heart rate and slight decrease in systolic blood pressure. The mean increase in heart rate in standing posture is 35 bpm. The mean decrease in systolic blood pressure is 9.3mmHg. The mean increase in diastolic blood pressure is 5mmHg.

Keywords: *Postural Orthostatic Tachycardia, orthostatic intolerance, Vitamin B12*

INTRODUCTION

In the last few years, there is an increase in the illness related to Autonomic nervous system. One of the subgroup of the disorders is the postural orthostatic tachycardia syndrome (POTS). POTS is characterised by orthostatic symptoms and dramatic increase in heart rate on standing, but does not involve orthostatic hypotension. Many a times,these patients with POTS may be misdiagnosed as anxiety neurosis, and potential therapeutic opportunities may be missed. In this article, we present and discuss a case of asymptomatic Postural Orthostatic tachycardia.

MATERIAL AND METHOD

A male student aged 19 years was examined routinely as a study volunteer. His height and weight were recorded. BMI was calculated. General physical examination was carried out. Then the subject was asked to lie down on the couch to rest for 10 minutes. Then his blood pressure was measured with Omron digital blood pressure monitor in different postures.

FINDINGS

The height and weight of the subject were 168.91 cms and 48 kg respectively. His Body mass Index (BMI) was 16.82kg/m^2. He is completely well and has no symptoms. He has no history of stressful situations like fever, trauma, and surgery and he is carrying out his daily activities comfortably.

The same subject was examined on three different days in the morning 2 hours after breakfast between 10:30am to 11:30am. On each day the subject was asked to lie down on the bed and rest for 10 minutes. The heart rate, systolic blood pressure (SBP) and diastolic blood pressure (DBP) were recorded in supine position with Omron digital blood pressure monitor. Then the heart rate, systolic blood pressure and diastolic blood pressure were recorded immediately after standing upright.

The data shows that there is marked increase in heart rate and decrease in systolic blood pressure and increase in diastolic blood pressure. The mean increase in heart rate is 35 bpm. The mean decrease in systolic blood pressure is 9.3mmHg. The mean increase in diastolic blood pressure is 5mmHg. We observed orthostatic postural tachycardia (mean increase in heart rate-35 beats per minute in standing posture) in him.

The table below shows the findings of the subject on three different days.

Table 1. Showing the Heart Rate, SBP, DBP of the subject on three different days

Day	Heart Rate(In bpm)			Systolic Blood Pressure(SBP)(In mmHg)			Diastolic Blood Pressure (DBP)(In mmHg)		
	Supine	Standing	Difference	Supine	Standing	Difference	Supine	Standing	Difference
1	69	100	+31	127	117	-10	60	68	+8
2	84	118	+34	116	110	-6	65	65	0
3	57	98	+41	120	108	-12	58	65	+7
Mean	70	105.34	+35	121	111.67	-9.3	61	66	+5

DISCUSSION

In the last two decades, there is a great increase in the understanding of illnesses related to autonomic disturbances. There is a subgroup of patients who suffer from distinct type of autonomic dysfunction manifested by orthostatic postural tachycardia. This disorder is known as Postural Orthostatic Tachycardia Syndrome (POTS) [1, 2].

Postural Orthostatic Tachycardia Syndrome (POTS) is defined as a condition in which marked rise in the heart rate of 30 beats per minute or greater occurring within 10 minutes of head up tilt or standing but without orthostatic hypotension[3].

The principle feature of POTS is orthostatic intolerance which produces symptoms on standing and these are relived on lying down. Patients with POTS have symptoms like palpitation, fatigue, light headedness, exercise intolerance, diminished concentration and syncope or near syncope. These patients have limitations to do normal activities of life like bathing, household work even eating food[4, 5].

At present, nearly 500,000 patients are affected by POTS in USA alone. Of this 25% are unable to work [6]. As the diagnosis is not readily made, the true prevalence may be likely to be higher [3]. Some patients with POTS may remain asymptomatic and escape the diagnosis.

The condition is common in adolescence and age group of 10-40 years, with female to male ratio of 5:1. Relatively very few people get affected after the age of 40 years and even in the young age group the symptoms resolve as the person grows older [3,7].

POTS is commonly seen after stress such as sepsis, fever surgery, trauma and pregnancy. POTS was first described by LOW et al. Robertson stated that it was one of the most common conditions in young females. The most common presentation involves the teenagers within the first three years of their growth spurt [8, 9].

Fu and colleagues have observed that upright heart rate and total peripheral resistance were greater, whereas stroke volume and cardiac output were smaller in patients than in controls. Left ventricular mass and blood volume were smaller in patients than in controls [10].

Oner and colleagues have observed in Turkish children that vitamin B12 is involved in the production of adrenaline and noradrenaline, catecholamine degradation and myelin synthesis. Significantly low vitamin B12 levels have been observed in patients with POTS, in adolescents compared to normal age matched subjects. During adolescence, the requirement of vitamin B12 increases because of increased myelin synthesis[11]. In POTS patients, a rise in plasma noradrenaline level in standing posture has been reported.

Most patients with POTS will improve over a period of time and they will be able to do their regular daily activities [12]. The subject in this study had lean body mass (LBM) of 33.908 kg and appendicular lean soft tissue ALST) of 22.422kg 1 3.

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To Study Seasonal Variation in Serum Total Testosterone Levels in Adult Males

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ABSTRACT

Levels of serum total testosterone vary with season. The levels of serum total testosterone have profound effect on metabolism of the body and affects weight and waist circumference. The present study was conducted to assess the seasonal changes in serum total testosterone levels. The study was conducted in 55 healthy male medical students of first professional M.B.B.S. and employees of 18 to 50 years age group at G.S.V.M. Medical College, Kanpur. The serum total testosterone levels were estimated by ELISA kit with the help of ELISA reader in the months of December & January and May & June. The change in mean weight, mean height, BMI, and Waist to Hip ratio of the subjects were found stastistically insignificant($p>0.05$). Waist circumference in Dec-Jan was 81.3±7.9 cm and in May-June was 80.6±8.0 cm. This decrease in waist circumference was significant ($p<0.05$). The serum total testosterone in Dec-Jan was 4.43±0.99 ng/ml and in May-June was 3.05±0.85. This decrease in serum total testosterone was significant ($p<0.05$).

Thus, it is concluded that in healthy men the serum total testosterone level and waist circumference vary with season, with higher level in the winter (Dec-Jan) and lower in summer (May-Jun).

Keywords: *Serum Total Testosterone, Waist Circumference*

INTRODUCTION

Testosterone is a steroid hormone from the androgen group. In mammals, testosterone is primarily secreted in the testes of males and the ovaries of females, although small amounts are also secreted by the adrenal glands. It is the principal male sex hormone and an anabolic steroid.

Various factors are believed to affect the testosterone levels including anxiety, intensity of exercise and amount of muscle mass utilized in the exercise, supplementation with oral zinc and calcium. Stressful environmental stimuli are thought to influence testosterone production rate and to reduce androgenecity [1]. Reduced secretion of testosterone with psychological stress and high altitude exercise. Other factors which influence testosterone levels are cortisol, testicular temperature, testicular blood flow and cholesterol.

Testosterone levels are highest in early twenties. The decrease in serum testosterone levels are now occurring at an even earlier age. Up to 50% of all men at 40 yrs now have testosterone level what was considered the normal range of 450ng/dl. Recent studies imply that the pesticides and preservatives in foods and hormone pellets to fatten up cattle, pork and chicken act as "hormone disruptors". Risks associated with low serum testosterone are cardiovascular diseases, stroke, prostate cancer, senile dementia and

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osteoporosis [2]. As the male body ages, gonadal function slowly declines with a resulting drop in serum testosterone of approximately 1% per year after age 30 [3].

MATERIAL AND METHOD

The present study was conducted in 55 healthy male medical students of first professional M.B.B.S. and employees of 18 to 50 years age group at G.S.V.M. Medical College, Kanpur. Subjects with a history of physical exercise, addiction, drug history, personal or family history of Diabetes, Hypertension and heart diseases, and history of any chronic disease were excluded from the study.

Anthropometric measurements: Height, weight, body mass index, waist circumference, and waist hip ratio.

Serum total testosterone level:

Collection of blood sample- 3 ml of venous blood was collected in the morning between 9-11 A.M. in the month of January and June at the Deptt. Of Physiology, G.S.V.M. Medical College, Kanpur, in a plain vial and allowed to clot and then centrifuged to separate the serum.

Method of Serum Testosterone Estimation

Serum total testosterone was measured by Enzyme-linked immune-sorbent assay (*ELISA*) at the Deptt. Of Pathology, G.S.V.M. Medical College, Kanpur.

The DRG Testosterone ELISA kit (Lot/Ch.B.: 29k091-2 & expiry date:30/09/2012 and Lot/Ch.B.: 29k121 & expiry date: 31/12/2012) manufactured by DRG Instruments GmbH, Germany a Division of DRG International Inc., USA was used for the quantitative in vitro diagnostic measurement of testosterone in serum. The reference values were:-

Population	5%Percentile	95%Percentile
Males	2.0ng/ml	6.9ng/ml

Statistical analysis

For each parameter, the mean and standard deviation were calculated in the months of Dec-Jan and May-June according to accepted methods. The mean difference of each parameter in both season were tested for significance by applying paired Students't'-test. All statistical tests were done by SPSS version 16.

OBSERVATIONS AND RESULTS

The present study was conducted on 55 healthy male medical students of first professional M.B.B.S. and employees of 18 to 50 years age with an average of 22.56±2.42 years in the Department of Physiology, G.S.V.M. Medical College, Kanpur. Estimation of serum total testosterone levels were done in the months of December, 2011-January, 2012 and May–June, 2012. The average environmental temperatures were 12°C in Dec-Jan (winter) and 40°C in May-June (summer).

The serum total testosterone in Dec-Jan was 4.43±0.99 ng/ml (Mean ± SD) and in May-June was 3.05±0.85 ng/ml (Mean ± SD). This Decrease in serum total testosterone was significant ($p<0.05$) (table-1).

Serum total testosterone

Table 1: Seasonal variation in serum total testosterone levels

Parameter	Dec-Jan Tests (n=55) Mean±SD*	May-Jun Tests(n=55) Mean±SD
Serum TT** (in ng/ml)	4.43±0.99	3.05±0.85

P<0.05

*standard deviation,

** serum total testosterone

Anthropometric measurements:

Table 2: Seasonal variation in Anthropometric parameters

Sl. No.	Parameters	Dec-Jan Tests (n=55) Mean±SD	May-Jun Tests(n=55) Mean±SD	p value
1.	Height (in mt)	1.69±0.06	1.69±0.06	>0.05(NS*)
2.	Weight (in kg)	62.37±8.91	62.59±8.75	>0.05(NS)
3.	BMI (in kg/m^2)	21.78±2.76	21.86±2.69	>0.05(NS)
4.	WC (in cm)	81.3±7.9	80.6±8.0	<0.05(S**)
5.	WHR	0.89±0.05	0.88±0.05	>0.05(NS)

*Not significant, **Significant

Waist circumference (WC) in Dec-Jan was 81.3±7.9 cm (Mean ± SD) and in May-June was 80.6±8.0 cm (Mean ± SD). This decrease in waist circumference was statistically significant ($p<0.05$) (table-2 & figure-2).

Other anthropometric parameters like weight, body mass index(BMI), waist hip ratio(WHR) were found insignificant ($p>0.05$) (table-2).

DISCUSSION

The present study was conducted on 55 healthy male first professional M.B.B.S. students and employees of 18 to 50 years age group. The aim of our study was to see seasonal variation in serum total testosterone levels.

Serum total testosterone

Serum total testosterone was significantly lower in the months May-June, 2012 in comparison to December, 2011-January, 2012. Our results coincide with the result of previous study of Svartberg J *et al* [4], who also found significant seasonal change in serum total testosterone, with a small peak in late winter (February), the lowest levels in summer (June through august) and a prominent peak in the fall (October and November), based on a cross sectional study of 1565 men, living in the municipality of Tromso, North Norway. Dabbs Jr JM [5] also reported a seasonal peak of serum total testosterone in December based on a cross-sectional study of 4462 U.S. military veterans aged 32-44 years. Reinberg A *et al* [6] also found a peak in November based on cross-sectional study of 207 subjects (participants in a pre-vasectomy study), living in Texas. But in a longitudinal study of five Parisian men, Reinberg A *et al* [7] reported a peak in total testosterone levels in October.

In contrast, Meriggiola MC *et al* [8] reported the highest levels of testosterone in May and June based on a longitudinal study of 16 healthy men. Nicolau GY *et al* [9], and Valero-Politi J *et al* [10] also found a peak of serum total testosterone in summer.

Although seasonality seems to be almost universal, its timing varies, possibly reflecting an effect of the duration of daylight or temperature on the reproductive system [11]. Kanpur (India) is located at 26.4670° North and 80.3500° East. Kanpur features an atypical version of a humid subtropical climate that resembles the climate of Delhi to some degree. Unlike many other cities with a humid subtropical climate, Kanpur features long and very hot summers, mild and relatively short winters, dust storms and a monsoon season. Kanpur lies in northern plains of India, which witness extremes of temperature. It can drop to a minimum of 0.0°C in the winters while it goes up to 48°C in summers. Kanpur experiences severe fog in December and January.

Svartberg J *et al* [4] explored the possibility that LH could be responsible for the seasonal pattern of testosterone, and observed a small seasonal variation for LH in Tromso. This was also observed by Nicolau GY *et al* [9], Reinberg A *et al* [6]. The seasonal changes in weight or physical activity also could be responsible for seasonal variation in serum total testosterone [4].

In our study, two subjects were married non-fathers who showed no difference in serum total testosterone levels when compared with unmarried males. In contrast, Peter B Gray *et al* [12] found that unmarried men had slightly higher levels of testosterone than the married non-fathers, but this difference was not statistically significant. Mathew McIntyre *et al* [(13)] also found that men who were romantically involved (i.e., are paired) have lower Testosterone than single men.

In our study, Waist circumference of subjects was significantly lower in May-June (summer) in comparison to Dec-Jan (winter). Similar findings were

noted by Visscher TLS and Seidell JC [14], reported that levels of BMI and Waist Circumference were lower in summer than in winter seasons. This decrease in Waist Circumference could be due to greater physical activity, smaller meal size and lesser rate of food intake in summer than in winter seasons. Uitenbroek DG [15] reported that people are generally more active in summer than in winter seasons.

Thus, it is concluded that in healthy men the serum total testosterone level and waist circumference vary with season, with higher level in the winter (Dec-Jan) and lower in summer (May-Jun). The seasonal changes in LH, weight or physical activity could be responsible for seasonal variation in serum total testosterone. The seasonal change in waist circumference could be due to change in physical activity and food intake behaviours.

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The Relationship between Body Fat Percentage and Pupil Cycle Time (PCT) in Healthy Indian Male of Various Nutritional Status

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ABSTRACT

Objective: To establish a relation between the body fat percentage (BF %) and Pupil cycle time (PCT) in healthy adult male.

Background: The Pupil Cycle time (PCT) is the parasympathetic response in the eye. This is a sensitive and specific test to determine the ophthalmological parasympathetic response. Cardiac parasympathetic tone is comparable with the ophthalmological parasympathetic tone.

Materials and Method: The PCT is calculated by throwing a beam of light on the edge of the pupil and measuring the time of a single constriction and dilatation of the pupil. Body fat percentage was calculated from the body weight and body density. Body density is calculated from log of 4-skin-fold thicknesses in different parts of the body.

Results: We obtained that the mean of fat percent in three groups was 14.2±4.47% in the undernourished, 19.49±5.61% in well-nourished and 29.3±2.13 in overweight/obese. In the present study, we established the fact that the PCT is altered according to the body fat percentage irrespective to their BMI.

Conclusion: It is established that there is a decrease in parasympathetic activity when there is more percentage of fat in body irrespective of their BMI and vice versa.

Keywords: *Pupil Cycle time (PCT), Parasympathetic nervous system (PNS), Body fat percentage (BF%), Skin-fold thickness, Body Density(BD)*

INTRODUCTION

Body fat percentage is an important indicator of health. Increase in fat percentage causes obesity and thereby type 2 diabetes, coronary heart disease, cerebrovascular diseases, high blood pressure[1]. The body fat percentage of a human or other living being is the total mass of fat divided by total body mass; body fat includes essential body fat and storage body fat[2]. Decrease in body fat also causes decreased immunity, decreased skeletal muscle function, productivity, increased morbidity ect[3,4]. Depressions in sympathetic and parasympathetic activity are significantly but weakly associated with increasing percentages of body fat. These associations indicate that in obese persons, autonomic changes though not necessarily causal, involve several organ systems including eye[5]. We suggest that autonomic alterations are important in human obesity and chachexia, as they are in animal obesity[6].

A disordered homeostatic mechanism may promote excessive storage of energy by decreasing

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sympathetic activity, while defending against weight gain by increasing parasympathetic activity. Stored body fat is used primarily as a reservoir for energy[5,7]. One of the biggest health problems in this country is excess fat due to overeating and decreased physical activity [8,9,10].

There are differences between essential fat and storage fat[1112,13,14,15]. We have intended to evaluate the effect of body fat percentage on pupil cycle time i.e parasympathetic response in the eye irrespective to their BMI or nutritional status.

MATEIRALS AND METHOD

Subjects

Sixty-four male adults in the age range of 18 to 50 years were included in the study. They were recruited from general population, male staff and student of St. John's Medical College. A brief history including a system review and clinical examination was performed. The person who had history of diabetes, hypertension, hypotension, asthma, other cardiac or ophthalmological diseases like cataract, optic neuritis, on chronic medication were excluded from the study. All of them were non-smoker, non-alcoholic and weight stable: those with noticeable weight gain or loss over the preceding six months were also excluded[16]. They were instructed to have their last meal at 8 pm, the night before and avoid caffeinated drinks for 12 hours prior to the study. They were divided in three groups according to their BMI as undernourished, wellnourished/normal and overweight/obese. Detail of the experimental procedure was outlined to the participants and an informed written consent was obtained. The ethics committee of St. John's Medical College approved the study[5,6,17,18,19].

Weight, height and BMI

Weight was measured to the nearest 0.01kg using an electronic scale(Afcoset HW – 100Ka1), in minimum clothing , and height was measured to the nearest 0.1 cm using a stadiometer(Raven Equipment Ltd, Dunmow, Essex, England) without sandals. Body mass index (BMI) was calculated by using Quetlet formula = body weight (kg)/ height^2 (m^2).

Skin folds thickness

Skin-fold thickness was measured by using Holtain calipers (Holtain Ltd, Crymych UK) which has standard pressure jaws of 10gm/mm^2. It was estimated to the nearest mm, except for a low values (usually 5 mm or less) when it is taken to the nearest 0.5mm. These reading were made at four sites on all subjects at the biceps, triceps, sub-scapular, and supra-iliac areas. The measurements were taken in triplicate by the same investigator on the right side of the body with the subject standing in a relaxed condition[17-22]. Durnin & Womersley calculation was considered valid for people between 17 and 68 years old [18,19,20].

Body Density

Body density is a measurement that expresses total body mass or weight relative to body volume or the amount of space or area your body occupies. Mathematically, it is body mass divided by body volume. Body density was determined by log of sum of four skin folds in mm and age and sex-specific formula[18,19].

Calculation of percentage of fat

Body composition was calculated by the body weight and four skin-fold thicknesses using age and sex specific formula to assess body density. Fat percentage was calculated using Siri's equation[18,19,20].

$$\text{Fat percentage (Fat\%)} = \left(\frac{4.95}{\text{Density}} - 4.5\right) \times 100$$

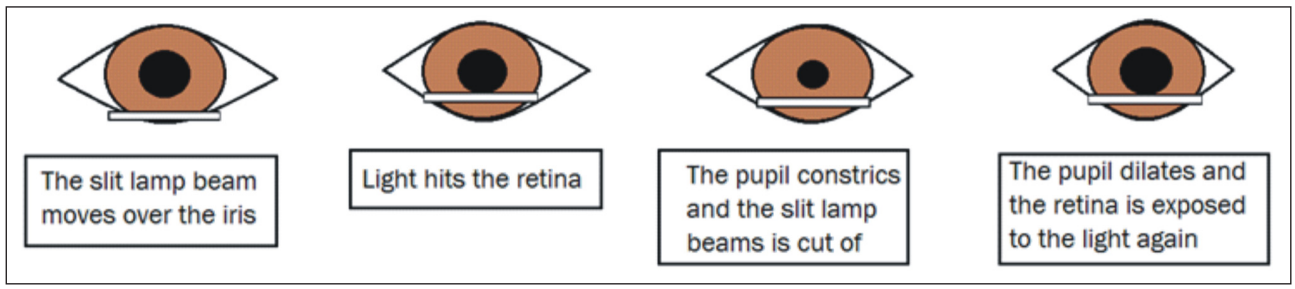
Slit –lamp

A Haggstreit-type of slit-lamp was used to measure the PCT, in the Department of ophthalmology at St. Johns' Medical College hospital. The same lamp was used for all the sixty four subjects studied. The subjects placed their chin on the chin rest. The height of the chin rest from the base of the table is adjusted in order to view the eye adequately for all the subjects with different heights. The slit beam was with horizontal axis and a thickness of 1mm as depicted in the picture below. The white diffuse horizontal light was used.

Measurement of Pupil Cycle Time (PCT)

The subject was seated for 15 minutes at the slit-lamp after entering the dimly light examination room to allow time for dark adaptation[23-26]. The slit lamp was adjusted to a comfortable height for both the subject and examiner. Then the subject was instructed to gaze to the imaginary infinity. The slit lamp was put on and the beam of the light was focused on the edge of the pupil of one eye from below. The constriction and dilatation of pupil started and that was counted for 30 cycles.

The subject was then rested for 15 minutes and the counts were repeated. Three sets of 30 cycles were noted.



The same procedure was followed for the other eye. If any problem like watering, too many blinking or illumination occurred during the measurement of any sets of 30 cycles, the entire reading was discarded.

The stopwatch has an oscillator of 1 kHz, which gives an accuracy of 1 millisecond. The counter started the stopwatch at the beginning and at the end of the preselected number of cycles; which reduced human error to a minimum. The PCT is measured in both the eyes[24]. The PCT which was of longer duration in the two eyes, was taken into consideration for analysis [28,29]. Data is rejected if there was blinking more than 2 times in one sitting or too much watering in the eye due to intensity of light[27,28,29].

Statistical analysis

Statistical analysis is performed by linear regression analysis with a P value of 0.05 and a confidence interval of 95%.

RESULTS

Data is expressed as mean±SD (Table 1) Our results show that Pupil cycle time (PCT) is directly proportional to the percentage of body fat (BF%) irrespective to the nutritional status(Fig1). The BMI is directly proportional to body fat (Fig 2). The body density (BD) is inversely proportional to the pupil cycle time (PCT) (Fig 3).

Table 1: Subject characteristics of three study groups with Pupil cycle time (PCT). Data is expressed as mean±SD

Subject characteristics	Undernourished BMI<18.5(kg/m²) N=22	Well nourished BMI=18.6-24.9(kg/m²) N=22	Overweight/ Obese>25(kg/m²) N=20
Age(yrs)	26±7.6	33±9.3	27±8.7
Height(cm)	1.63±0.05	1.67±0.06	1.68±0.47
Weight(kg)	44.9±4.87	59.63±7.46	88.7±12.1
BMI(kg/m²)	16.7±0.02	21.2±1.83	31±4
Fat mass(% of body wt)	14.2±4.47	19.47±5.61	29±2.13
Density(gm/cm³)	1.065±0.010	1.052±0.011	1.029±0.007
PCT(millisecs)	818±145	904±63	991±101

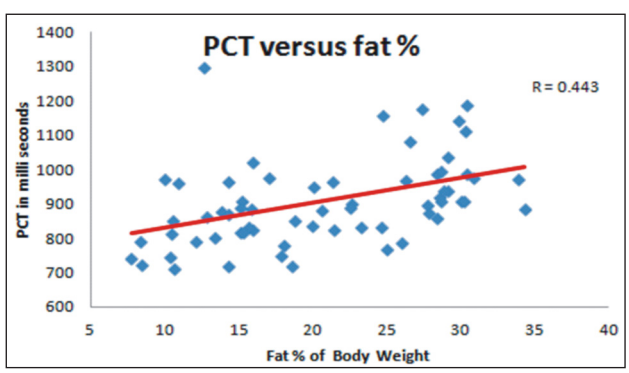


Fig. 1. A graph of statistical fitment is carried out through linear regression analysis to establish a relation between PCT and BF% in all three groups namely undernourished (14.2±4.47), well-nourished/normal (19.47±5.61) and overweight/obese .

DISCUSSION

In this cross-sectional study of clinically healthy male with stable weights, we found that the activity of human parasympathetic nervous system in the eye was inversely correlated with the percentage of body fat. Anthropometry is generally considered the single most easily obtainable, inexpensive and noninvasive method that reflects body composition[18]. We have calculated the BF% using log of sum of four skin folds in mm and age and sex-specific formula to determine the body density using Siri's equation. The mean %fat in three groups were 14.2±4.47 in undernourished, 19.49±5.61 in well-nourished, and 29.3±2.13 in overweight/

obese(Table 1). There was increase of fat percentage with increase of BMI although not well demarcated between undernourished and well-nourished group as shown in some other study[19,20]. A direct correlation between BF% and PCT was obtained which shows parasympathetic response in the eye is decreased with increase of BF% irrespective to their BMI with an r value of 0.44 (Fig 1).

We have also obtained a relationship between PCT and BD which is inversely proportional to each other as it was expected. In the three groups the BD remained as 1.065±0.010 in undernourished, 1.052±0.011 in well nourished and 1.029±0.007 in overweight/obese. There was no significant difference between the first two groups but there was obvious difference between well-nourished and overweight/obese; undernourished and overweight/obese. A correlation line could be drawn in the three groups with an r value of -0.522.

Our results are not likely to be explained by proved or suspected confounders of autonomic indices like diet[6], acute weight change[21], sex[3], some common exposures(for example to ethanol and caffeine), sodium depletion as well as illness and medications were addressed in our entry criteria of experimental protoco[5]. Since women were not enrolled, we do not know whether our findings applied to them[23]. There have been some reports that physical activity and fitness influences autonomic indexes even in the resting stage, although this has not been a consistent Some studies reflects the idea that percentage of body fat and autonomic nervous system are not limited to a single organ system[15,24]. Contrary to other study, our study suggests that there was an increase in the parasympathetic activity in the eye when there was increase in fat percentage[5,6]. In the light of our findings, it is important to pursue studies that simultaneously examine both the sympathetic and parasympathetic components of the autonomic nervous system in various body composition irrespective to the nutritional status like unnourished, well nourished(normal), or overweight/obese. Such studies would determine whether profiles of autonomic nervous system can be used to characterize clinically important subgroups of overweight persons or the undernourished.

Our study has also established that there is a strong correlation between body fat and BMI with an r value of 0.79. As other study suggest we also obtained a steady increase of body fat percentage with increase of BMI[24]. Some individuals who are overweight are not overfat. Others have BMIs within the normal range and yet have a high percentage of their body weights as fat[11]. In this study, we also observed that these misclassified persons are uncommon relative to the population as a whole.

CONCLUSION

We conclude, our study demonstrates that there was a decrease of parasympathetic activity in the eye with an increase of body fat percentage irrespective of their nutritional status (undernourished, well-nourished or overweight/obese). There was an increase of fat percentage with increase of BMI. The parasympathetic response in the eye is decreased i.e increase in PCT with decrease in body density as evidenced in the increase of body fat.

Limitation of the study: The study did not investigate the sympathetic activity. Therefore, evaluation of the sympathetic activity in the eye in different fat percentages in order to assess the response of both sympathetic and parasympathetic influence with regards to function of iris is necessary. This study also did not adjust the subject with their physical activity level.

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Conflict of Interest: We declare that there is no conflict of interest.

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Prevalence of Helicobacter Pylori in Asymptomatic Paediatrics Patients in a Tertiary Hospital : A Cross-Sectional Study

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ABSTRACT

The present hospital based cross sectional study was done on 214 paediatric patients attending O.P.D for symptoms other than those of gastrointestinal disorders and the prevalence of H.pylori was found using Stool Ag card test. Out of these 214 patients,112 were found positive for H.pylori by stool antigen card test giving a prevalence of 52.34.%. A detailed proforma was filled having information regarding age and sex of the patient, education and occupation of the head of the family, sanitary practices, dietary habits of the patient was also observed for pallor. Among a total of 117 males, 64 were positive for H.pylori (54.7%) and among 97 females, 48 were positive (49.5%). Age wise distribution showed maximum prevalence of H.pylori in the age group of 15-18 years (75.9%) and minimum in the age group of 3-6 years (30.4%). A higher prevalence of H.pylori was found among non-vegetarians (53.1%) and among patients having pallor (75.8%) which was significant. A study of socio-economic status showed a prevalence of H.pylori as 52% in upper lower and 52.6% in lower middle socio-economic group.

Keywords: *H.Pylori, Stool Antigen Card Test, Pallor*

INTRODUCTION

Helicobacter pylori is a gram negative curved, microphilic and motile organism. It is a common bacterium infecting about half the world's population[1]. There is substantial evidence that it causes chronic gastritis peptic ulcers, duodenal ulcer and is also involved in development of gastric carcinoma[2-4] Actual infection rates vary from nation to nation with developing world having higher rates than developed countries[1,2].The age at which this bacterium is acquired seems to influence the possible pathologic outcome of the infection. Infections are usually acquired in early childhood in all countries[5]. Acquisition at an older age brings different gastric changes more likely to lead to duodenal ulcer[6]. High rates of seropsitivity in children are found in many developing countries[7]. However, in developing countries the prevalence is higher and as much as up to 90% figure has been reported[8,9]. Once acquired, H. pylori infection generally persists for life, unless treated by specific antimicrobial therapy.[8]H.pylori consist of a large diversity of strains and the genomes of three have been completely sequenced[10-14]. Occupationally acquired infections have also been reported especially among, endoscopists and gastroenterologists[8,12-14].Another possible route is faecooral and H, pylori has been isolated from faeces of infected young children[16,17]. There has also been studied investigating the association between the seroprevalence of H. pylori and Hepatitis A virus[18-23]. Consumption of uncooked vegetables irrigated with

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water contaminated with untreated sewage was associated with H. pylori seropositivity[24]. The municipal water supply has greater chances of spreading H. pylori infection as compared to private water supply[25]. Various socio-economic conditions comprising of high density crowding, poor sanitary practices family's income, educational level and occupation[26-28] have been held responsible in spreading of pathogens..Early detection of H. pylori population and its eradication may result in a significant improvement in severity of dyspeptic symptoms. It is important to find out H. pylori prevalence and identify high risk population so that treatment strategies can be appropriately planned.

Hence the present cross sectional hospital based study was done on patients attending OPD for conditions other than gastro-intestinal disorders and the prevalence of H. pylori was estimated. Other parameters like socio-economic status according to Kuppuswamy grading, age, sex, diet, pallor were also studied and association of H. pylori, if any was seen.

MATERIAL AND METHOD

The study was conducted in the department of Paediatrics at GSVM Medical College, Kanpur in collaboration with department of Physiology at KGMU, Lucknow during 2012-2013. A total of 214 patients of both the sexes attending OPD for symptoms other than those of gastrointestinal disorders were screened for H.pylori. Written informed consent was taken from parents of all the patients after explaining them the nature and purpose of study. Ethical clearance was taken prior to the study from the ethical committee. Patients who had taken proton pump inhibitors or antibiotic for a month prior to study were excluded from the study. Patients stool sample was collected in an airtight container and the stool assay was performed using Immunocard STAT HpSA test. (Standard diagnostics Inc). HpSA test is a non-invasive and an accurate test especially useful for screening of asymptomatic subjects. Pallor was seen in the lower palpebral conjunctiva.

Statistical Analysis: Data were analysed by the chi-square test to compare the association between different variables and H.pylori positive rates. A value of $P < 0.05$ was considered statistically significant. The calculations were done using the software package SPSS 16.0.

RESULTS

Out of total 214 patients, 112 patients were H.pylori positive by Immunocard STAT HpSA test, giving a hospital based prevalence of 52.34% (Table1). Out of total 117 males, 64 were positive for H.pylori (54.7%) whereas out of 97 females 48 were positive (49.5%) (Table 2). The prevalence was estimated in different age groups. The maximum number of positive patients was found in the age group of 15-18 years (75.9%) and the minimum prevalence was in the age group of 3-6 years (30.4%) (Table 3). For socio economic status, the groups were classified according to modified Kuppuswamy scale for urban families. Out of total 175 patients belonging to upper lower group 91 were positive for H.pylori (52%), 38 belonging to lower middle socioeconomic group, 20 were positive (52.6%) and only one child of upper middle group was found and was positive.(Table 4). Out of total 214 patients, 96 were non-vegetarians with 51 positive for H.pylori (53.1 %) and 118 were vegetarians with 61 positive (51.7%) (Table 5).Pallor was present in 62 patients with 47 positive (75.8 %) and absent in 152 patients with 65 positive for H.pylori (42.8%) (Table 6).

Table 1: Prevalence of H.pylori

Total subjects	214
Subjects positive with H.pylori card test	112

Prevalence of H.pylori == 112/214 X 100 =52.34 %

Table 2: Number of H.pylori positive patients according to sex

Sex	Total subjects	subjects positive for H.pylori	Percentage %
Males	117	64	54.7%
Female	97	48	49.5%

Table 3: Number of H.pylori positive patients according to age group

age group (years)	Total subjects	No. of positive patients	Percentage %
3-6	46	14	30.4 %
7-10	74	40	54.1 %
11-14	65	36	55.4 %
15-18	29	22	75.9%

Table 4: Number of H.pylori positive patients according to socio economic status

SE status	Total subjects	No. of positive patients	Percentage %
Upper lower	175	91	52 %
Lower middle	38	20	52.6 %
Upper middle	1	1	100 %

Table 5: Number of H.pylori positive patients according to diet

Type of diet	Total subjects	No. of positive patients	Percentage %
Vegetarian	118	61	51.7%
Non- Vegetarian	96	51	53.1%

Table 6: Number of H.pylori positive patients according to pallor

Pallor	Total subjects	No. of positive patients	Percentage %
Present	62	47	75.8 %
Absent	152	65	42.8 %

DISCUSSION

This study was carried to find out the prevalence of H.pylori among patients attending OPD for symptoms other than gastrointestinal disorders. These patients were screened for H.pylori by Immunocard STAT HpSA test. In a study from South India, 105 children were screened for H.pylori and its prevalence rate varied from 44% to 46%[27]. The overall prevalence recorded in our study was 52.34% which is higher in comparison to the above study. This can be explained by the fact that prevalence of H.pylori varies widely by geographic area, age, race, and ethnicity and SE status. In our study, age wise distribution showed maximum prevalence in the age group of 15-18 years (75.9%) and minimum in the age group of 3-6 years (30.4%). A study from Hyderabad has shown that H.pylori infection increases with age with 60% at 3-10 years, 50% at 11-15 and 84% by 16-20 years of age[28].In a study from Mumbai, it has been shown that the prevalence of IgG antibody was 22%, 56% and 87% in 0-4, 5-9 and 10-19 years age group respectively in 340 subject[29]. Another similar study from Bangalore has detected H.pylori infection in 82% of 50 children of 6-18 years of age by ^{13}C urea breath test[30].Kang etal in a report from South India found 57% of subjects between 6 months to 4 years positive for IgG antibodies for H.pylori[31].Sharma etal too reported a 50% seropositivity for H.pylori in children below 10 years of age[32]. In a study from Chennai in an urban upper class population, a 21.1% prevalence rate was seen in individuals between 12-20 years of age[33]. In the present study, among H.pylori positive patients 54.7% were males and 49.5% were females. Although there is a slightly greater male preponderance but the difference between the genders was not significant which is similar in the study from South India[27].In the present study, out of 62 patients having pallor 47 patients were positive for H.pylori (75.8%) which is significant.H.Pylori colonization appears to impair iron uptake and increase iron loss. Regarding the possible role of H.pylori in iron deficiency anaemia, a recent metaanalysis indicated that the infection is associated with depleted iron deposits[34]. A study from Bangladesh has shown the prevalence of iron deficiency anaemia with decrease in haemoglobin and serum ferritin was significantly higher in H.pylori infected patients[35]. The prevalence of H.pylori in our study was found to be higher in low socioeconomic groups being 52% in upper lower and 52.6% in lower middle groups. This is consistent with previous studies which have demonstrated that the prevalence of H.pylori as well as gastritis is more frequent in those who come from large families, have poor hygiene, low standards of living, poor sanitation practices and overcrowded living conditions[36-38]. Socioeconomic status is not restricted to income and social class but also considers other factors such as living standards, urbanization and educational level[39]. A prevalence of 51.7% was seen in vegetarians and 53.1% in non-vegetarian group which was though higher in non-vegetarians but was not significant ($p>0.05$), which supports the fact that it is probably the food prepared under unhygienic conditions which plays a role in transmission of H.pylori in developing countries and not the type of food consumed[40].

CONCLUSION

The present study revealed substantial prevalence of H.pylori in asymptomatic children with males more affected and maximum prevalence in the age group of 15-18 years. The prevalence is higher in low socioeconomic classes with poor sanitation practices and unhygienic water supply. A higher prevalence of H.pylori was seen in subjects having pallor and non-vegetarians which may be the contributing factor in the development of peptic ulcer and gastric cancer. In conclusion, identification of the populations, who do not show symptoms of H.pylori infection but still harbour it, is essential for controlling the infection and it still remains a challenge for clinicians.

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A Comparative Study of Lipid Profile in Smokers and Non Smokers in Bareilly Region

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ABSTRACT

Background: Smoking is one of the environmental factors which can alter normal lipid profile. It is one of the major risk factors in the genesis of coronary atherosclerosis and development of coronary heart disease.

AIMS: To comparatively evaluate the lipid profile alteration in smokers (>5years smoking) and non smokers.

Material and Method: The study was conducted on 672 randomly selected apparently healthy male subjects of age group 20-60yrs, of these 336 were smokers and 336 nonsmokers. The subjects selection criteria was on the basis of voluntary participation The subjects were asked to fast overnight, and early morning blood samples collected and analyzed for lipid profile by appropriate standard methods. The student's unpaired "t" test was used for statistical analysis. P-value of (< 0.05) was considered statistically significant.

Results: It was noted that mean S.TC (202.83 ± 25.63 mg/dl),and LDL-C (120.59 ± 29..91 mg/dl), were significantly higher in smokers (p <0.05) as compared to non-smokers, i.e., mean S.TC 196.91 ± 21.71mg/dl), LDL-C (111.01± 31.91). Mean serum TG levels in smokers were also significantly high, i.e., 153.71 ± 21.1 mg/dl (p <0.001) as compared to non-smokers with mean serum TG levels (148.19 ± 22.28 mg/dl). Mean serum HDLC(37.42 ± 10.41 mg/dl) was significantly lower in chronic smokers (p <0.05) as compared to non-smokers, i.e., 38.82 ±7.95 mg/dl.

Conclusions: Smoking causes alterations in lipid profile. Serum total cholesterol, triglyceride, LDL-C were significantly increased, while serum of HDL-C was low in smokers in comparison to non-smokers. This alteration in serum lipid levels increases risk of coronary artery disease.

Keywords: *Smokers, Non-Smokers, Lipid Profile Parameters*

INTRODUCTION

Cigarette/Bidi smoking is the most common type of tobacco use .Cigarette smoking is socially accepted in various communities and has drastically increase around the world[1]. The World Health Organization reported that tobacco smoking killed 100 million people worldwide in the 20th century and warned that it could kill one billion people around the world in the 21st century[2]. By the early 2030, tobacco related deaths would increase to about 10 million a year[3]. Cigarette smoking is one of the leading causes of preventable morbidity and mortality, and is one of the major single preventable cause of ill health in the world. Cigarette smoking is one of the major cause of pulmonary and cardiovascular diseases in developing and developed countries. Cigarette smoke contain an important chemical substance nicotine. It is highly addictive psychoactive chemical. Nicotine stimulates sympathetic adrenal system leading to increased secretion of catecholamine resulting in increased

lipolysis and increased concentration of plasma free fatty acids (FFA) which further results in increased secretion of hepatic FFAs and hepatic triglycerides along with VLDL-C in the blood stream[4-5]. These changes contribute to the atherosclerotic potential of cigarette smoke. Most of the studies concluded that smokers have higher levels of total cholesterol (TC) , triglyceride (TG), atherogenic VLDL-C, low density lipoprotein (LDL-C) with lower level of antiatherogenic high density lipoprotein cholesterol (HDL-C) in the pathogenesis of atherosclerosis.[6] Cigarette smoke contains approximately 10^{17} oxidant molecules per puff that can cause damage to lipids, proteins, DNA, carbohydrates, and other biomolecules[7] . It is becoming increasingly evident that a prooxidant/ antioxidant imbalance largely contributes to atherosclerosis processes.[8] It has been postulated that many of the adverse effect of smoking may result from oxidative damage to critical biological substances.[9] Previous reports have demonstrated abnormal endothelial function in chronic smokers.[10] Endothelial dysfunction in turn has been proposed to play a pathogenic role in the initiation of vascular disease.[11] Although smoking induced endothelial dysfunction is very likely multifactorial more recent clinical and experimental observations strongly point to a potential role of oxygen derived free radicals in mediating this phenomenon

There are conflicting reports regarding the lipid profile differences between smokers and nonsmokers. The alteration in lipid profile parameters-total cholesterol (TC), triglyceride (TG), HDL-C, LDL-C, will be evaluated in a group of smokers and will be compare with the data from non smokers. The present study is an attempt to find out the extent of these differences in a sample population from U.P., India. This would help in arriving at a more firm conclusion.

MATERIAL AND METHOD

This was an observational population based cross-sectional study comprising of 672 randomly selected male participants (336 smokers and 336 non-smokers) of age group 20-60years. This study was conducted in the Physiology department in association with Department of Biochemistry of Rohilkhand medical college & Hospital, Bareilly, U.P., during the period 2011-2013. 336 male smokers between the ages of 20 to 60 years who were smoking for more than 5 years were included, only males were considered because in our society males smoke more frequently and openly compared with females. For the comparative assessment, 336 healthy individuals (non smokers) served as control. The inclusion criteria of study include smokers who smoke more than 10 cigrettes/ bidis for more than 5 years who were non-alcoholic and non-obese. The exclusion criteria includes subjects < 20 yrs and >60yrs of age, obese, persons with angina, diabetics, alcoholics, subjects with renal failure, hepatic failure, family history of lipid disorderes, tuberculosis or other chronic respiratory illness and females. All subjects were age and BMI matched and were of almost same socioeconomic status. Detailed history, name, age, occupation, personal history and personal habits of the subjects were recorded. Smoking history was noted in detail. These were noted in a personal Performa and parameters concerned with the study were recorded. Written informed consent from all the participants were taken and clearance from institutional ethical committee was taken. The lipid profile parameters include serum Total Cholesterol [TC], Triglycerides [TG], Low Density Lipoproteins [LDL-C], High Density Lipoproteins [HDL-C]. The subjects were asked to fast overnight and early morning blood sample from the antecubital vein of each subject [5 ml blood] was collected under aseptic precautions. Samples were processed within 1 hour for quantitative lipoprotein cholesterol measurements using the vertical spin ultracentrifugation technique. Serum was obtained by centrifugation for 4 min at 3000 rpm and was then transferred into properly labeled sterile vials and stored at - 20°C till the determination of lipid profile. Serum TC was measured by CHOD-PAP method[12], serum TG by GPO -TRINDER method[13] and HDL-C tests by Phosphotungstic Acid precipitation method[14] whereas LDL-C was determined by calculation method.[15] All the tests were done on ERBA Chem-5 semi autoanalyser within 1 hour after collection of sample in biochemistry laboratory.

RESULT

In this study 672 male participants (336 smokers and 336 non-smokers) were screened. Table-1 Shows that both the groups (smokers and Non-smokers) were comparable in age, body weight, height and body mass index (BMI) as their differences in mean were not statistically significant (p>.05)

Table 2 Shows the observed values for fasting lipid profile among smokers and non-smokers.

A statistically significant increase in TG, (p=0.001), TC (p=0.001), LDL-C (p=0.0001) and statistically significant decrease in HDL-C, (p=.05) was observed in smokers as compared to non-smokers.

Table 1: Showing physical characteristics (Mean ± SD) in smokers and Non-smokers

Parameters	Smokers n=336	Non smokers n=336	P Value
Age in years	36.32±11.03	35.54 ±10.59	.355
Body weight (Kg)	58.20±4.07	58.44±4.36	.454
Height (Cm)	166.36±9.13	167.17±9.21	.255
BMI (Kg/m2)	21.24±2.99	21.14±3.24	.666

Table 2: Showing lipid profile in smokers and Non-smokers (Mean ± SD)

Test	Smokers n=336	Non smokers n=336	P Value
TG(mg/dl)	153.71 ± 21.10	148.191 ± 22.28	.001
TC(mg/dl)	202.83± 25.63	196.91 ± 21.71	.001
HDL-C(mg/dl)	37.42 ± 10.41	38.82 ± 7/95	.05
LDL-C(mg/dl)	120.59 ± 29.91	111.01 ±31.91	.0001

Comparative.

'P'<0.05 was considered significant.

Statistical Analysis

The whole data was analyzed using SPSS version17 software. The test applied was unpaired't' test to see the difference between the groups, if any

DISCUSSION

Atherosclerosis is the underlying process involved in coronary artery disease, peripheral vascular disease and stroke[16].Smoking causes a huge and increasing number of untimely death in India[17]. Tobacco smoke contains many constituents; nicotine is one of the main constituents. Nicotine and other toxic substances from tobacco smoke are absorbed through the lungs into the blood stream and are circulated throughout the body. These substances narrow or damage the blood vessel walls; hence plaques form at a faster rate in smokers.[18] There is definite relationship between tobacco use and atherosclerosis. Cigarette smoking adversely alters the serum lipid and lipoprotein levels.[19,20,21] This study was conducted to evaluate the effect of smoking on lipid profile. The current study showed significantly higher levels of TC, TG, LDL-C and significantly decreased serum HDL-C levels in smokers as compared to non- smokers. The change in serum lipoprotein levels are more marked in chronic smokers. The increased TC, TG, LDL-C levels were also observed in the study of Friedman[22]. The findings have also been substantiated by Neki.[23] and Akbari[24] et al. Statistically significant increase in TG, TC, LDL-C levels and decrease in HDL-C levels were also in accordance with the result found by Afroz Afshan et al[25]. Kesaneimi and Grundy[26] also found statistically significant increase in LDL-C levels in smokers than non-smokers, but Nesje et al[27] found no significant difference between smokers and non-smokers concerning TG, and TC levels. Similarly no significant alterations were observed by Raya et al[28].

The most prominent feature in our study lies in the level of anti-atherogenic HDL-C, which showed a significant decrease in smokers (p<.05) as compared to non-smokers and this observation was supported by Dussa et al[29]. In agreement with our finding, a fall in HDL-C levels by 3-5 mg/dl in smokers has previously been reported by Rosenson[30]. Contrary to the our findings Dirican et al[31] did not find significant differences in serum TC, TG, LDL, HDL levels between smokers and nonsmokers. Dyslipidemia is a well-established risk factor for the development of coronary artery disease. Our study demonstrated presence of dyslipidemia in chronic smokers. Increased levels of TG, TC, LDL-C, and decreased levels of HDL-C in smokers will facilitate to development of coronary artery disease. HDL is an antiatherogenic substance; its fall increases the risk of cardiovascular diseases.

CONCLUSION

Smoking causes dyslipidemia and this dyslipidemic state lead to an increased risk of coronary artery disease. The policies that prevent and reduce smoking will have positive response and large benefits for decreasing cardiovascular mortality[32]. Creating awareness regarding health consequences of smoking in schools and colleges by establishment of mandatory public health education and other antismoking advice should be made an important part of public health system.

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A Study of Variation in Heart Rate Variability with Change in Posture in Young Adult Indian Females

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ABSTRACT

Context: Heart rate variability (HRV) is an important measure of autonomic functioning, which provides powerful means of observing the interplay between the sympathetic and parasympathetic nervous systems. In normal women, there was found predominance of parasympathetic over sympathetic tone. The HRV response to postural change is a sensitive measure of the shift in autonomic balance from parasympathetic to sympathetic predominance that, when attenuated or absent, has been correlated with prevalent disease in patients.

Aim: The purpose of the present study was to systematically investigate the effect of change in posture on HRV parameters using frequency domain measures in young adult Indian females.

Settings and Design: The study was conducted on fifty young adult females.

Method and Material: Height in centimetres (cms), body weight in kilograms (kgs), Pulse and BP was noted.HRV was recorded with the help of Anu photo rheograph by frequency domain method in three postures in a sequence: lying, sitting and standing.

Statistical analysis used: SPSS 17.0, one way ANOVA and Tuckey's Post Hoc test were used to find out statistical significance of the results.

Results: The values of the mean RR Interval and mean HF for females in lying, sitting and standing positions were 0.75 ± 0.13, 0.74 ± 0.11 and 0.66 ± 0.08 and 26.46 ± 16.59, 21.25 ± 8.21 and 14.89 ± 8.11respectively. Both decreased significantly when posture changed from supine to sitting to standing.

The values of the mean LF for females in lying, sitting and standing positions were 20.20 ± 8.37, 31.11 ± 9.59 and 28.44 ± 12.19 respectively. It increased significantly when posture changed from supine to standing to sitting.

The values of the mean TP and mean VLF in three positions were not significantly different.

Conclusions: The decrease of mean RR interval and mean HFand an increase of mean LF with changes in posture shows that females have higher cardiac parasympathetic modulation and lesser sympathetic modulation which contributes as one of the cardio-protective mechanisms in them.

Keywords: *Heart Rate Variability, Posture Change, Autonomic Nervous System, Young Adult Females, Indian*

INTRODUCTION

Measures of heart rate variability (HRV) are a reliable reflection of many physiological factors modulating the normal rhythm of the heart.[1] In fact; they provide a powerful means of observing the interplay between the sympathetic & parasympathetic nervous systems. A growing number of studies indicate that increased variability in the heart's interbeat interval is physiologically desirable.[2]A depressed heart rate variability level usually indicates the presence of pathological conditions such as coronary artery disease, heart failure, diabetes and

hypertension. HRV is also a predictor of left ventricular dysfunction following myocardial infarction and is a risk factor for morbidity and mortality.[1] HRV is important because it provides a window to observe the heart's ability to respond to normal regulatory impulses that affect its rhythm. A heart response time to the sympathetic stimulation is relatively slow. Upon stimulation of the sympathetic nervous system it takes about 5 seconds to start increasing HR and almost 30 seconds to reach its peak level. A heart response to the parasympathetic stimulation is almost instantaneous. Depending on actual phase of heart cycle, it takes just 1 or 2 heartbeats before heart slows down to its minimum level proportional to the level of stimulation. At rest, both sympathetic and parasympathetic systems are active with moderate parasympathetic dominance. The actual balance between them is constantly changing, maintaining an optimum body function.

In addition to the autonomic nervous system (ANS), the external factors, like body posture also change the spectral characteristics of HRV. In the supine position, the parasympathetic modulation is dominant, and causes stronger high-frequency heartbeat fluctuations. In contrast, decreased parasympathetic function occurs in the standing position.[3]The HRV response to postural change is a sensitive measure of the shift in autonomic balance from parasympathetic to sympathetic predominance that, when attenuated or absent, has been correlated with prevalent disease in patient populations.[4]

The numerous recorded observations on the effects of posture on the pulse-rate deal mainly with the different rates in the lying, sitting, and standing positions. So far as explanations have been offered of postural effects on pulse rate and blood pressure, these have been attributed to hydrostatic influences affecting the amount of blood in the splanchnic area and the blood pressure in the head, in addition to the different amounts of static muscular contraction needed to maintain the different postures.

The present study was aimed to evaluate the difference in HRV parameters due to postural changes in young adult females. One of the best ways to assess the autonomic function is to analyze minute changes in heart rate, which are caused by many factors including regulatory influence of the ANS. HRV can be assessed by time domain or frequency domain indices.

Frequency domain measures of HRV provide information on the frequency distribution of the components of HRV using power spectral density analysis.

The high frequency (HF) component (0.15Hz -0.40 Hz) measures the influence of the vagus nerve in modulating the Sinuatrial node and the inspiratory inhibition of the vagal tone.

The low frequency (LF) component (0.04Hz-0.15 Hz) influenced baroreceptor-mediated regulation of blood pressure and reflects predominantly sympathetic activity.

The very low frequency (VLF) component (0.003Hz -0.04 Hz) reflects the influence of several factors on the heart, including chemo receptors, thermo receptors, the Renin-angiotensin system, and other non-regular factors.

The purpose of the present study was to systematically investigate the effect of change in posture (lying to sitting to standing) on HRV and compare various components of HRV in the three postures in healthy young adult females in India.

MATERIALS AND METHOD

A total of 50 healthy young adult female volunteers were included in this study with age range from 18 to 25 years.

The study was conducted in the Department of Physiology at a Medical College.

The non smoker, non alcoholic, non diabetic, having normal pulse rate, blood pressure, normal heart sounds and having no evidence of illness and having perfect physical, mental and psychological well being were included in the study.

A brief history was taken and general physical examination of all the volunteers was done with main emphasis on cardiovascular diseases, renal diseases. None of the subjects took any medication at the time of study. All the tests were carried out between 11 am to 4 pm. The procedure was explained and informed consent was obtained after the subjects had read a description of the experimental protocol, which was approved by the ethical committee of the college. The height, weight and blood pressure of the subject was measured with measuring tape, weighing machine and sphygmomanometer respectively. On auscultation, the heart sounds were found to be normal.

Height was measured in centimeters (cms), body weight was measured in kilograms (kgs).HRV was recorded with the help of Anu photo rheograph using traditional frequency domain method. The experiment consisted of 3 recordings, each performed in a sequence: supine, sitting and standing positions. he experiment consisted of 3 recordings and each performed in a sequence: lying position, sitting position and standing position. During the data collection, the volunteers were instructed not to speak or move. To evaluate the autonomic HR modulation response in relation to the supine, standing and sitting postures, data were recorded for a 5-minute period at rest for each condition respectively, with spontaneous breathing. Initially the subject was asked to lie down over a bench in horizontal supine position and relax. The probe of pulse oxymeter was clipped to the subject's index finger and care was taken that subject did not move his hand. The probe was connected to the Anu-photo-rheograph which was in turn connected to personal computer with application software (Variability Analyzer 2008). Record in lying position was taken.

After the first record, the subject was asked to get up and sit in a chair with hands placed on the bench at the level of her thorax and the probe of pulse oxymeter was attached to the index finger. Subject was asked to relax and record in sitting position was taken. At last the subject was asked to stand up with hands by the side of the body and the record was taken in standing position.

The recorded HRV raw data was analyzed in the frequency domain to get HRV graph and FFT power spectrum. Very low frequency (VLF), low frequency (LF), high frequency (HF) spectral powers were determined by integrating power spectrum between 0.00-0.04 Hz, 0.04-0.15 Hz and 0.15-0.4 Hz respectively and expressed in normalized units (nu). Total power was calculated between 0.00-0.5 Hz and expressed in absolute unit of millisecond squared.

The data was entered into and analyzed using SPSS 17.0 statistical package. The results were displayed using descriptive statistics. Further comparison between groups (supine, sitting and standing) was done using one way ANOVA followed by Tuckey's POST HOC test. The statistical analysis of the significance on the data was done using independent t-test. The statistical significance level was established at 5% ($p < 0.05$) and 1% ($p < 0.01$).

RESULTS

Table1. Physical Characteristics of Female Subjects

Gender	Physical Characteristics		
	Statistics	Parameter	
	Age	Height	Weight
Female			
Mean	19.12	156.21	54.86
Std. Deviation	2.04	22.71	7.00

Table 2. Descriptive Statistics and One Way Anova

Parameter	Posture	Mean	Standard Deviation	F-stat	DF	P value
Mean RR Interval	Lying	0.75	0.13	12.983	2,147	0.000*
	Sitting	0.74	0.11			
	Standing	0.66	0.08			
Total Power	Lying	1483.00	1759.75	0.562	2,147	0.571
	Sitting	1360.63	684.34			
	Standing	1235.69	746.22			
Very Low Frequency	Lying	18.20	13.79	0.269	2,147	0.764
	Sitting	18.06	7.79			
	Standing	16.83	8.41			
Low Frequency	Lying	20.20	8.37	15.480	2,147	0.000*
	Sitting	31.11	9.59			
	Standing	28.44	12.19			
High Frequency	Lying	26.46	16.59	12.470	2,147	0.000*
	Sitting	21.25	8.21			
	Standing	14.89	8.11			

*: Statistically significant at 1%level of significance i.e. P-value < 0.01

Table3. Multiple Comparisons (Tucky's HSD)

Parameter	Posture	Posture	Mean Difference	P- value
Mean RR Interval	Lying	Sitting	0.01361	0.795
		Standing	0.09810*	0.000*
	Sitting	Lying	-0.01361	0.795
		Standing	0.08449*	0.000*
	Standing	Lying	-0.09810*	0.000*
		Sitting	-0.08449*	0.000*
Total Power	Lying	Sitting	122.36735	0.862
		Standing	247.31373	0.540
	Sitting	Lying	-122.36735	0.862
		Standing	124.94638	0.855
	Standing	Lying	-247.31373	0.540
		Sitting	-124.94638	0.855
Very Low Frequency	Lying	Sitting	0.14319	0.997
		Standing	1.37482	0.783
	Sitting	Lying	-0.14319	0.997
		Standing	1.23163	0.823
	Standing	Lying	-1.37482	0.783
		Sitting	-1.23163	0.823
Low Frequency	Lying	Sitting	-10.90886*	0.000*
		Standing	-8.24299*	0.000*
	Sitting	Lying	10.90886*	0.000*
		Standing	2.66587	0.393
	Standing	Lying	8.24299*	0.000*
		Sitting	-2.66587	0.393
High Frequency	Lying	Sitting	5.20641	0.071
		Standing	11.57180*	0.000*
	Sitting	Lying	-5.20641	0.071
		Standing	6.36539#	0.019
	Standing	Lying	-11.57180*	0.000*
		Sitting	-6.36539#	0.019

#: Statistically significant at 5%level of significance i.e. P-value < 0.05

*: Statistically significant at 1%level of significance i.e. P-value <0.01

The mean RR Interval for females in lying, sitting and standing was 0.75 ± 0.13, 0.74 ± 0.11 and 0.66 ± 0.08 respectively. It is significantly different in three postures (F (2,147) = 12.983, p = 0.000).

The mean LF for females in lying, sitting and standing was 20.20 ± 8.37, 31.11 ± 9.59 and 28.44 ± 12.19 respectively. It is significantly different in three postures (F (2,147) = 15.480, p = 0.000).

The mean HF for females in lying, sitting and standing was 26.46 ± 16.59, 21.25 ± 8.21 and 14.89 ± 8.11respectively. it is significantly different in three postures(F (2,147) = 12.470, p =0.000).

The mean total power (TP) for females in lying, sitting and standing was 1483.00 ± 1759.75, 1360.63 ± 684.34 and 1235.69 ± 476.22 respectively. It is not significantly different (F (2,147) = 0.562, p> 0.05).

The mean VLF for females in lying, sitting and standing was 18.20 ± 13.79, 18.06 ± 7.79 and 16.83 ± 8.41 respectively. It is not significantly different (F (2,147) = 0.269, p > 0.05).

The mean difference in RR Interval for females between lying and standing and sitting and standing was 0.0.9810 and 0.08449{significantly different (p = 0.000)}. The mean difference in RR Interval for females between lying and sitting was 0.01361{not significantly different (p > 0.05)}.

The mean difference in LF for females between sitting and lying and standing and lying was 10.90886

and 8.24299 {significantly different (p = 0.000)}. The mean difference in LF for females between sitting and standing was 2.66587{not significantly different (p > 0.05)}.

The mean difference in HF for females between lying and standing and sitting and standing was 11.57180 and 6.36539{significantly different (p=0.01and p=0.019 respectively)}.The mean difference in HF for females between lying and sitting was 5.20641{not significantly different (p > 0.05)}.

Conclusion - With changes in posture (supine-sitting-standing), decrease of mean RR interval was observed. The result shows that the mean RR interval for females in three postures was significantly different (p=0.000). Sitting-standing difference of mean RR interval showed significant decrease. This finding is explained by increase in sympathetic tone with posture change from sitting to standing.

The mean LF for females in three postures was significantly different (p < 0.01). There was a significant increase of LF component with change in posture from lying to sitting. But with change of posture from sitting to standing, LF decreased not significantly. These findings can be explained by increase of sympathetic tone with lying-sitting change in posture. And decrease of sympathetic influence with sitting-standing change in posture could be related to recovery process trying to find a balance in a new standing condition. With changes in posture (supine-sitting-standing), decrease of HF was observed. The mean HF for females in three postures was significantly different (p = 0.000). When postures were changing from lying to sitting and to standing, there was decrease of HF component. This correlates with decrease of parasympathetic tone and increase of sympathetic influence with postural changes from lying to sitting and to standing positions.TP component of HRV decreased with postural changes but not significantly. In our study no significant difference in VLF component could be observed with change in posture. The mean difference in TP and VLF were not significantly different.

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Effect of Treadmill Exercise on Blood Parameters of Healthy Population

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ABSTRACT

Background: Exercise & physical activity is an important function of living system. It may affect Haematoligcal parameters and brings alternation in the interior setting. The objectives of the present study were to examine & compare the blood parameters of the pre & post treadmill exercise.

Method: This study was carried out at the Dept of Physiology, Navodaya medical college, Raichur. Fifty healthy subjects were included in the study with age group of 17 -23 yrs. The standard test protocol was 30 mints jogging. Study was performed with voluntary participation of healthy male subjects of our campus. Venous blood sample drawn from the group before & after exercise (30 mints jogging) on treadmill. The blood is investigated for the changes in Hb, Total WBC. This worked started after being confirmed by ethical committee of the college.

Results: The mean age was 22.8 years. Immediately after termination of exercise the Hb & Total WBC levels were increased significantly ($p<0.01$) when compared to Pre-exercised values.

Conclusion: It is concluded that exercise is physiological stress to body which is healthy. It induces certain changes that enhance the ability to cope with stress. Physiological training can considerably alter related changes in blood that may relate to training-induced hormonal influences remains to be workout.

Keywords: *Blood parameters, Exercise, Stress, Hb, Leucocytes*

INTRODUCTION

Blood is a complex fluid and has some specific dynamics of its circulation. Physical fitness is an active state that enables a person to do everyday activities without being easily tired, participate in leisure activities enthusiastically and overcome difficult situations[1]. Blood is a liquid tissue that its main function is to maintain the steady state of internal environment of body tissues or, in other words, to keep the homeostasis [2]. Like other organs of the body, blood does not have the same response to any physical activity. Type, intensity, time, and duration of activity are involved in the response of body to different activities[3]. Exercise is an important function of living systems. It affects many systems in our body. Many observations have shown that blood composition changes as a result of exercise. Various kinds of exercise were reported from first three decades of this century which dealt with differential blood counts of peripheral blood[4]. Nowadays public exercise, especially morning exercise, specially jogging and working out on treadmill, is popular among different groups of people due to its ease and convenience[5]. Blood viscosity is reduced after exercise and even more after regular training[6-8]. These effects result in an increased blood fluidity in regularly exercising individuals, which contrasts to the hyper viscosity syndrome of sedentary people. Clearly, while inactivity is characterized by a downward spiral in all physiologic functions, reflected by an increased viscosity, an improvement of blood rheology occurs parallel with the correction of

metabolic and body composition disturbances when people are submitted to training. These observations are potentially important since blood viscosity is now considered to play a role in cardiovascular risk[9-11]. In 1953 Rohde and Wachholder were the first to describe that leukocyte count showed a significant increase to almost maximal values with the first minute of exercise[12]. During the late 70's there were a number of studies, showing that blood increasing number of investigations about the acute immune response to exercise. The number of evaluations during the past years grew a lot[13-17]. The number of circulation of white blood cells and platelets by dynamic physical activities in humans can rapidly increase[18]. However, debates on the effects of exercise on hematological parameters still continue. While some studies indicate an increase in hematological parameters after intensive training exercise, some other studies show that hematological parameters after exercise did not change significantly. The aim of present study was to examine & compare the blood parameters of the pre & post treadmill exercise.

MATERIAL AND METHOD

The study was carried out at Department of Physiology, Navodaya Medical College, Raichur. To study this fact, the blood parameters of subjects of fifty healthy subjects of age group between 17–23 years, were examined for haematological investigations before and after exercise. The venous blood was collected and used for all haematological investigations. The sample of blood was collected by1.2 mg of anhydrous salt of EDTA per ml. of blood was used as an anticoagulant[13]. The standard test protocol i.e. 30 mints jogging was used. Study was performed with voluntary participation of healthy male subjects of our campus. Venous blood sample drawn from the group before & after exercise (30 mints jogging) on threadmill. The blood is investigated for the changes in Hb and Total WBC. Haemoglobin (Hb) was estimated by Sahli's haemometer, using cyanomethaemoglobin method, while improved Neubauer chamber was used for Total Leukocyte Counts (TLC). Blood smears were prepared and blood films stained with Leishman's stain for Differential Leukocyte Counts (DLC). Values obtained after exercise were compared with those observed from before exercise.

RESULT

The mean age of male subjects was found to be 22.8 years. Table-1 compares the blood parameters before and after exercise in male subjects. The result shows that, there was statistically significant increase in hemoglobin value (p<o.o1) after treadmill exercise. WBC was also higher than before exercise values and difference were statistically significant (p<0.001).

Table 1: Effect of exercise on blood parameters of male subjects before and after exercise (Mean ± SD)

Parameters	Before exercise	After exercise	Mean difference	t-value	p-value
Hb (g/dl)	14.9 ± 0.75	15.92 ± 0.59	1.01	11.82	P<0.001
WBC (10^3 / µL)	6.60 ± 0.43	7.18 ± 0.44	0.58	14.21	P<0.001

DISCUSSION

It is being accounted that exercise put forth physiological 'stress' on the body and hence there were number of hormonal and cellular changes, alongside physical change as elevated blood pressure, body temperature and oxygen intake. This depends on various factors as body status, nutrition, type and duration of exercise, etc. Exercise also induces immune like response, results leukocytosis that is quantitatively equivalent to the response against physiological insults to immune system.

The leucocytosis of exercise has been often compared to tenderness like reaction. As exercise induced in percentage of reactive leucocytosis is considered to be dependent on various factors, we have found that after strenuous exercise the number of leukocyte become almost double and the response is different among the subject as indicated by a large SD. This variability is most probably a result of difference in the physical body status and nutrition. The exercise induced leucocytosis has been often compared to an inflammation like reaction.[19, 20]

Leukocyte count of 12,000–20,000 per cubic mm during infections are not unusual and it is comparable to our mean WBC counts which raised up to 15,000 after exercise. The discussion that, improvement of systemic cortical levels due to exercise induced an inflammation like response is also supported by the

fact that the raised level of cytokine results increase secretion of adernocorticotrophic hormone. It suggests that, hormones of this regulatory circle are known to increase by the exercise. Principally monocytes and thrombocytes are responsible for the initiation of exercise induced acute phase reaction. The main source of cytokines belonging into TNF family and of IL–1 are regular monocytes.[21-23] Mature monocytes have a lower capacity to produce these cytokines. Release abroad spectrum of cytokines by regular monocytes and activated macrophages.

And it is the second wave of cytokines release that augmented homeostatic signal and initiates the cellular and cytokines cascades that are involved in the complex process of exercise induced, acute phase reaction. Being highly chemotactic for neutrophils, e.g., (IL–8) and mononuclear cells, result molecules emerge from local tissue site.[24]

Physiological studies have shown that stress from any source can influence on the endocrine, haemopoietic and immune systems. Cytokines and Cortisol seems to play an important role in the communication between these systems. The well-documented changes that occur are increase in erythrocytes, neutrophils and platelets, whereas lymphocytes, eosinophils and monocytes decrease in number.

CONCLUSION

Our finding suggest that physical stress ropes to treat by increasing WBC count and combating infections hence normalize the homeostasis and minimize dose and duration of medication. It induces certain changes that enhance the ability to cope with stress. Physiological training can considerably alter related changes in blood that may relate to training-induced hormonal influences remains to be workout. Recovering from illness can be monitored by the white cell count by the rise and fall indicating the improvement or worsening the condition.

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Ethical Clearance: The study was approved by Institutional ethical committee.

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Comparative Study of Neonatal Serum Bilirubin Level after Spontaneous Versus Oxytocin Induced Labour

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ABSTRACT

Objective: To investigate and compare the neonatal bilirubin levels in oxytocin induced delivery and spontaneous vaginal delivery.

Materials and Method: Eighty full term parturients were selected for this study .All had uncomplicated pregnancies and were under no medications except for iron preparation . The sub jects were divided into two subgroups according to onset of labour and mode of delivery. The first group consisted of 30 healthy babies of women who had received oxytocin infusion during labour for induction and the second group which formed the control group consisted of 50 healthy babies of women with normal vaginal delivery following spontaneous onset of labour

Neonatal serum bilirubin was measured on in cord blood on day 1 and later on capillary blood bilirubin level was measured on 3rd and 5thday of delivery and the datas were compared

Results: The levels of bilirubin in oxytocin induced group was significantly higher than those of spontaneously delivered group on day 1 and day3 (1.366±0.2563 versus 1.1662±0.3091, P=0.0039) & (5.719 ± 0.7624 versus 5.2703 ± 0.9497,P=0.0482) while the levels were higher but not significantly so on day 5 (4.661 ± 0.5663 versus 4.4976 ± 1.017 , P=0.4452)

Conclusion: Induction of labour with oxytocin does not seem to cause neonatal hyperbilirubinaemia or neonatal jaundice.

Keywords: *Oxytocin, Induction, Neonatal Bilirubin*

INTRODUCTION

Hyperbilirubinaemia is one of the most common causes of health problems, observed in 60% of term and 80% of preterm infants in the first week of life [1]. It is manifested with bilirubin level of more than 5 mg/dL and is harmless in some cases but may lead to neurotoxicity in severe condition Therefore early detection and treatment of neonatal hyperbilirubinemia is crucial in the prevention of bilirubin induced encephalopathy .[2,3]

There are many factors which can lead to development of hyperbilirubinaemia in a neonate. One of the factor cited by many authors is the liberal use of oxytocin for inducing labor [4,5,6]. Other reasons include immaturity of glucuronyl transferase enzyme, blood group incompatibility, certain drugs used by the mother and abnormal deliveries .

Some studies have also suggested about the dose dependent effect of oxytocin on the level of bilirubin in cord blood [7] It is well known that when oxytocin is

administered by continuous IV infusion, it results in expansion of maternal extracellular fluid (ECF) and consequently of the fetal ECF due to their constant transplancental equilibrium , by virtue of its antidiuretic effects. As a result the erythrocytes swell and become osmotically more fragile. These swollen and hyperfragile erythrocytes are easily trapped by the spleen, resulting in high bilirubin level[4]Another possible explaination could be enhanced placento-fetal transfusion due to oxytocin-induced uterine contractions, with resultant increase in red cell mass in neonates .[4]

Inspite of these findings, oxytocin infusions is still commonly used for induction of labour often without any medical necessity. This widespread liberal use of oxytocin prompted us to investigate and compare the effect of oxytocin and neonatal bilirubin levels with spontaneous vaginal delivery.

So the present study was done with an aim to determine the ability to predict severe hyperbilirubinemia in term healthy newborns delivered through oxytocin induced labour.

MATERIALS AND METHOD

Eighty full term parturients were selected for this study darbhanga medical college and hospital. All had uncomplicated pregnancies and were under no medications except for iron preparation . The subjects were divided into two subgroups according to onset of labour and mode of delivery. The first group consisted of 30 healthy babies of women who had received oxytocin infusion during labour for induction and the second group consisting of 50 healthy babies of women with normal vaginal delivery following spontaneous onset of labour formed the control group . All the gestation were of 38 weeks duration or more .None of newborn infants had any signs of RDS. Newborn infant who were growth retarded or born with APGAR score of less than 6 were all excluded from the study .Umbilical cord was clamped within 3 minutes of birth and all the babies were breast fed. Approval was obtained from the Institutional Ethical committee and all the participants gave written informed consent. Bilirubin was measured on day1 ,3 and 5 after delivery. About 10 ml of blood samples were collected from umbilical cord from the placental site of the divided umbilical cord cord for day 1 measurement. Later on ,neonatal capillary blood was obtained by heel prick on day 3 and 5 under strict aseptic precautions.Bilirubin was measured by spectrophotometry

All datas are expressed as mean ±standard deviation (S .D) Statistical analyses was done using graph pad instat software .Unpaired t test was used for comparison between dose of oxytocin used for induction of labour and the serum bilirubin levels obtained on the three days Statistical significance was accepted at $P<0.05$.

RESULT

The datas of 30 neonates in group 1 who were born via vaginal route after labour induction with oxytocin and 50 neonates in group 2 (control group) who had normal spontaneous delivery without oxytocin infusion were analysed. The indication for induction were mainly rupture of the membranes and oligohydromnios beyond 39 weeks of gestation The base line characteristicsof the groups were presented in table 1. The difference in number of parity and labour was statistically significant ($p<0.001$) .Mean age and parity of subject in group 1 and group 2 was 22.7±4.219 years & 26.38±5.283 years and 1.4±0.4983 & 2.8775±0.5997 respectively with significant difference between them. Mean gestational ages of the groups were similar. The bilirubin levels in oxytocin induced group 1were significantly higher than those in control group 2 (spontaneous delivery) on day 1 and day 3 (1.366±.2563 versus 1.1662±o.3091, P=0.0039)and (5.719±0.7624 versus 5.2703±0.9497, P=0.0482) respectively while the levels were higher but not significantly so on day 5 (4.661±0.5663 versus 4.4976±1.017 , P=0.4452)

Correlation between the doses of oxytocin used for induction and the level of bilirubin on all the 3 days was assessed and no significant correlation was found.

Table 1: The baseline characteristics of the groups

Parameters	Group 1(n=30) Mean ± Sd	Group2 (N=50) Mean± S.d	P
Maternal Age	22.7± 4.219	26.38± 5.283	0.0023
Gravida	1.4±0.4983	2.8775±0.5997	<.0001
Gestational weeks	39.6667±0.6609	39.10±0.8391	0.0023
Type of delivery	Induced with oxytocin	Spontaneous	
Male gender ratio	60%	54%	
Oxytocin dose (units)	6.35±2.429	0.00	

Table 2: The distribution of metabolic parameters between the groups

Parameters(newborn Bilirubin)	Group 1 (N=30) Mean ± S.d	Group 2 (N=50) Mean± S.d	P	Significance (p < 0.05)
DAY 1	1.366±0.2563	1.1662±0.3091	0.0039	SIGNIFICANT
DAY3	5.719±0.7624	5.2703±0.9497	.0482	SIGNIFICANT
DAY5	4.661±0.5663	4.4976±1.017	0.4452	INSIGNIFICANT

DISCUSSION

Various studies on neonatal bilirubin levels and the use of oxytocin for the management of labour have produced conflicting results but it has been widely accepted that oxytocin infusion during labour increased the risk of neonatal hyperbilirubinaemia [6,8,9,10,11,12].Our findings on day 1 and day 3 are consistent with these studies. However some other recent studies have not shown any association between oxytocin administered to the mother during labour and serum bilirubin levels in infants [13,14,15] .Our findings on day 5 was in agreement with these studies.

In our present study the level of serum bilirubin levels in group1 were significantly higher than those in group 2 on day 1 and day 3 (P=0.0039,P=0.0482) respectively while the levels were higher but not significantly so on day 5 (P=0.4452)

However the levels of serum bilirubin of the present study is within the normal limits as bilirubin levels normally rises to 5-10 mg/dl by the 3rd to 4 th days of neonatal life and decreases thereafter . [16]

The elevated bilirubin levels were only of biochemical and not of clinical concern and none of the babies during this period received any phototherapy or any other medical treatment or both.

Several theories have been reported to explain the higher bilirubin level after induction with oxytocin .

Singhi et al in his study had explained that oxytocin when administered by continuous IV infusion, results in expansion of maternal ECF with dilutional hyponatraemia and hypo-osmolality by virtue of its antidiuretic effect .

Since maternal and fetal body fluids are in constant transplacental equilibrium, an expansion of fetal ECF occurs, as a result erythrocytes swell and become osmotically more fragile. These swollen and hyperfragile erythrocytes are easily trapped by the spleen, resulting in higher bilirubin.[4]

A relatively immature glucuronyl transferase system due to absence of the hormonal upsurge of normal labour[10] and an enhanced placento-fetal transfusion due to oxytocin-induced uterine contractions, with resultant increase in red cell mass in neonates , have also been suggested.[17]

Other mechanism are trauma to the fetal erythrocyte as a result of uterine activation ,vasoconstrictive effects of oxytocin on uterine blood vessel and hyponatraemia caused by the the administration of large quantities of electrolyte free diluents for oxytocin infusion [5,14,18] .

Howevever studies done by Seidmann et al , oppose these assumptions [5] .

Linn et al also reported oxytocin did not affect neonatal bilirubin levels [19].

Maissels et al reported that breastfeeding and the percentage of weight loss after birth were major determinants for the neonatal jaundice rather than oxytocin infusion in the healthy newborns [20].

The findings of the present study were a bit different from the previous ones as there was significant

increase on serum bilirubin in oxytocin induced newborns on day1 and3 but it was not so on day 5 . However,it could be due to the fact that in our study 5% glucose solution was used as a diluents for oxytocin . Omigbodun et al proposed that 5%dextrose used as diluents for oxytocin increase the risk of transplacental hyponatraemia due to the infusion of large volumes of salt free fluid into the mother and neonatal hypoglycaemia and neonatal hyperbilirubinaemia as a consequence[18]. So the increase in serum bilirubin levels on day 1 and 3in the present series could be due to use of 5%dextrose as a diluent for oxytocin rather than oxytocin itself.

Despite our utmost efforts , we might have missed on any events that may have affected the result .Though we did not observe any factors that may have lead to high bilirubin level in the neonatal period.

Most importantly our results could be an incidental findings as study group was not very large. Further studies are required for more clarity.

CONCLUSION

We found no difference between the labour inductions with

with the above doses of oxytocin and that of spontaneous delivery in properly selected cases in regards to neonatal hyperbilirubinaemia or neonatal jaundice as the increase in serum bilirubin in induced cases were only of biochemical concern and not a clinical problem

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Biochemistry of ABO Blood Groups: A Review

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ABSTRACT

The ABO blood group polymorphism of humans are known to be determined by the expression of A, B or H (O) antigens which are terminal neutral glycan sequences found in abundance on glycoprotein's and glycolipids . Apart from their matching role in blood transfusion these specific antigens are known to play their important role in regulation of various biochemical and immunological reactions in the body and are with platelets as intrinsic determinants and extrinsically absorbed antigens and exist both on glycosphingolipids and glycoprotein's . Throughout the years there have been many reviews of the ABO blood group system, covering different aspects of this fascinating topic. This article provides a brief introduction to this amazing complex blood group system and our intension is to focus on the biochemistry of ABO blood groups.

Keywords: *ABO Blood Groups, Glycoprotein's, Glycosyl Transferase*

INTRODUCTION

Blood groups are genetically transmitted biochemical expressions with antigenic (agglutinogen) properties located within red blood cells. They tend to provoke antigen antibody reaction in presence of a specific antibody (agglutinin) in plasma. The process of agglutination can also be provoked by adding blood of one with another species. The antigen-antibody reaction is known as agglutination which results in aggregation of red blood cells followed by haemolysis and release of hemoglobin. A short resume therefore of the relevant theory both of antigen and antibodies and genetics is a useful starting point for an introduction to human blood group system in terms of the ABO and Rh system.

Historical background

The ABO blood group system is widely credited to have been discovered by the Austrian scientist Karl Landsteiner, who found three different blood types. Landsteiner described A, B and O groups [(1)]. Decastrello and Sturli discovered the fourth type AB [(2)]. The ABO system is the most common and important antigenic system in red blood cells and has great relevance for blood transfusion in humans. An individual's ABO blood group was early used in paternity disputes by Lawyers, in the study of different populations by anthropologist, in forensic science by police. Two types of agglutinogens are present in the red blood cells which are called as A agglutinogen and B agglutinogen. If A group has two subgroups 'A_1' and 'A_2' similarly 'AB' group has two subgroups namely 'A_1B' and 'A_2B'. Individuals belonging to group A have A antigen in the red blood cells and b-antibodies and B group people have B antigen in the red blood cells and a-antibodies in the plasma. Those belonging to blood group AB have both A and B antigen and no antibody (α or β). They are the universal recipient. Group O have no reactive antigens but have both a and b antibodies in the plasma and called as universal donors.

Biochemistry of ABO blood groups

Biochemically the defining sugars in blood groups A, B, AB and O are nitrogenous substance, neutral

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heteropolysaccharide containing D-galactose, methyl pentose fucose, D-glucosamine, D-galactosamine present as N-acetyl derivatives and other amino acids such as threonine, serine etc. [3,4]. Each substance has however its own very definite specificity and it depends upon the terminal components Which may be non-reducing sugars and was taken as a sign of large precursor molecules carrying small residues that differentiate blood groups[5]. It is possible to study the structure which determines the specificity and the way in which the blood group gene operate to produce these specific substances.

According to theories of inheritance genetic material consists of DNA which is arranged in such way as to provide information of amino acid for a given protein. This would mean that proteins are the only direct result of gene action. The difference between A, B, H substance does not however, seem to be in their protein structure but in the carbohydrate portion of the molecule. This suggest that blood group substances are not the direct product of the gene but that the blood group gene produce protein enzyme which act at late stage upon basic mucopolysaccharide to give blood group specificity [6]. It is suggested that a common blood group substance exists which is converted though a series of stages to the final specific blood group products. The conversions are almost certainly affected by specific enzymes the synthesis of which is controlled be genes [7]. The ABO locus is located on chromosome 9 at 9q34.1- 9q34.2. The A and B allele differ from each other by 7 nucleotide substitution, four of which translate into different amino acids in the gene product.

The A or B specificity of the glycosyl transferase can be determined by the residues present at position 266 and 268 .The almost entirely different protein lacking enzymatic activity in O allele are the result of deletion of guanine at position 261 causes a frame shift and translation and differ from the A allele[8] .

The biochemistry of the A and B antigens was elucidated by the astonishingly early and brilliant work from the group of Kabat; 1956 and Morgan and Watkins; 1953[4, 9]. The A, B and H determinants were hypothesized to reside on water-soluble glycoprotein able to inhibit agglutination of RBCs by antibodies or lectins A precursor substance H was hypothesized as a building structure for A and B and the terms O – (or H-) substance and anti H were introduced by Morgan and Watkins in 1948 .

An H gene codes for a fucose transferase that imparts, a fucose on the end of the glycolipid or glycoprotein, forming the H antigen that is usually present in individuals of all blood type [10]. The H locus is found on chromosome 19 at 19q13.3 and it encodes a fucosyl transferase that synthesizes the H antigen on RBCs.

The basic structure of antigen H is given below.

↓

Fucose-Gal-GalNAC – Protein

The presence of A and B glycosyl transferase was first predicted by Watkins in 1967 and then experimentally established by other workers [11, 12] .

Individuals who are type A have a gene which code for a transferase (N-acetyl-galactosaminyl transferase) that catalyze placement of a terminal N-acetylgalactosamine on the H-antigen where as individuals who are type B have a gene which codes for a transferase (galactosyl transferase) that place a terminal galactose [13].

A antigen : Fucose Gal NAc – Protein

↓

GalNAc

B antigen: Fucose – Gal-GalNAc – Protein

↓

Gal

Individuals who are type AB have both the transferases and who are type O have neither, so the H antigen persists.

Thus A and B glycosyl transferase use UDP-Gal NAc and UDP Gal respectively as substrates [14] .Both

require the H determinant as acceptor. The O protein is nonfunctional leaving the H-defining terminal fucose unaltered. The Secretor (Se) locus is found on chromosome 19 at 19q13.3 and encodes a Fucosyl transferase expressed in the epithelia of secretory tissues like salivary glands, the gastrointestinal tract and catalyzes the synthesis of H antigen in bodily secretions and A and B antigens.

There genotype is Se/Se or Se/se. The genotype of nonsecretors is se/se and are homozygous and are Unable to produce H antigen and hence cannot be processed into A and B antigens. If H gene absent no H substance can be formed, and therefore no A or B antigen (extremely rare). Result is Bombay blood group. The Bombay blood group lacks H gene and therefore cannot make H antigen (H substance). Since the H substance is the precursor for the A and B antigens, these antigens also are not made. The cells

Type as O and the serum has anti-A, anti-B, and anti-H since the individual lacks all of these antigens anti-H agglutinates O cells. The only cells Bombay individuals do not agglutinate are from other Bombay blood people since they lack the H antigen.

The ABH sugars are found on glycolipids (approximately 10percent) and glycoproteins (approximately 90 percent) on the RBC as well as on many different tissues and cell types ,including epithelial cells that line the lumen of the gastrointestinal, respiratory ,and reproductive tracts as well as in salivary glands and skin .The term histo-blood group often being used to reflect this wide distribution. A and B antigen synthesis occurs during normal glycosylation of proteins and lipids in the Golgi compartment .The precursor H substance is synthesized by one of the two fucosyltransferases depending on the acceptor substrate used .The FUT1 gene that encodes the 2-alpha fucosyltransferase is responsible mainly for the synthesis of the H antigen on type 2 (type 4) carbohydrate precursors found on RBCs[15] .The closely related FUT2 gene encodes a very similar 2-Alpha fucosyltransferase (FucT) that is expressed in epithelial cells and synthesizes H antigen mainly on type 1 and type 3 chains. Because H antigen is the precursor for both A and B antigens neither antigen can be synthesized without alpha Fuc T activity, independent of the ABO genotype.The A and B glycosyltransferases are type II membrane proteins located in the Golgi compartment[16,17] although soluble forms are found in plasma and other body fluids .The enzyme consists of a short transmembrane domain, a stem region , and a catalytic domain that extends in to the Golgi lumen.

Patenaude and colleagues described the crystal structure and showed that the catalytic site is divided in to two domains; the N-terminal domain recognizes the nucleotide sugar donor substrate (UDP-Gal or UDP-GalNAc), whereas the acceptor substrate is held by the C- terminal domain .Manganese ion is essential to the reaction because of its ability to bind the UDP part of the donor substrate, lies between the two domains. In addition two so called disordered loops have been identified by the crystal structural studies. One is located at the C-terminal of the enzyme .The other disordered loop lies close to the catalytic site of the enzyme. Mutations in this region have been shown to reduce enzyme activity and are often associated with weak subgroup phenotypes [18]

Antibodies of the System

Anti-A and Anti –B antibodies are naturally occurring antibodies that are produced by immunocompetent individuals from the age of approximately 6 months .The widely held dogma is that these antibodies are in fact mimicking antibodies produced against terminal carbohydrates on bacterial cell wall as a response to our normal intestinal microbial flora, and that these glycotopes share structural homology with A and B antigens. Anti-A and Anti-B are predominantly IgM antibodies although IgG or IgA components are often found [19, 20]. Pathogenic interactions (Bacteria ,viruses, and parasites) have been proposed as important driving forces for the geographic distribution of the ABO blood group phenotypes because different pathogens demonstrate blood group –identical or –cross reactive molecules on their surfaces .These are the probable targets for "blood group " antibodies , and their existence is the leading hypothesis as to why we make naturally occurring antibodies against the carbohydrates blood groups we lack .In addition many pathogens show selective binding to blood group carbohydrate moieties via lectins [21].Of all the antibodies to RBC blood group antigens , anti – A and anti-B are the most clinically important and its significance extends beyond transfusion medicine and is also important in both organ and hematopoietic transplantation.

The Rhesus System

It is the second most important blood type system in the human blood transfusion Landsteiner and Wiener discovered the Rh blood group antigen or Rh haemogen in 1940 in the red blood cell of certain individuals. It is named after the Rhesus monkey, because it was used in experiments which finally culminated in the discovery of this system. The red blood corpuscles are agglutinated by the ant rhesus sera obtained by injecting the red blood cells of rhesus monkey (the common brown monkey Macaques rhesus of India) into rabbits or guinea pigs. Such

individuals are said to be Rh-positive while others whose red blood cells lack the Rh antigen are known as Rh-negative. The Rh antigen is present at birth and is inherited as a single Mendelian dominant by a pair of allelomorphic genes Rh and rh. The terms Rh-positive and Rh-negative which are in clinical use are regarded as synonyms with 'D' positive and 'D' negative respectively. The discovery of Rh factor has been proven to be of value in determining the paternity of a child.

CONCLUSION

The ABO blood group antigens appear to have been important throughout our evolution because the frequencies of different ABO blood types vary among different populations, suggesting that a particular blood group conferred a selective advantage. People with common blood group O express neither A nor B antigen, and they are perfectly healthy. Numerous associations have been made between particular ABO phenotype and an increased susceptibility to disease.

Use of powerful new techniques of genetics, genomics and proteomics integrated with system physiology of different blood group individuals with the special interest in the glycoprotein's of blood group 'A' and 'B' (N-acetylgalactosamine and galactose) may be helpful in early diagnosis , proper cure and management of diseases occurring in particular blood group persons .

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