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Effect of Bhramari Pranayama Practice on Cognitive Functions in Healthy Volunteers

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Abstract

Bhramari Pranayama (Humming bee breath) is one of the common slow pranayama practices, which involves inhaling through both nostrils and while exhaling produce sound of humming bee. Practicing pranayama reduces the effects of stress and strain on various systems by increasing the vagal tone. With the ongoing COVID-19 pandemic, there are heightened feelings of stress, anxiety and depression and pranayama can be an effective way to improve our mental and emotional well-being. This study has been chosen to assess the effect of Bhramari pranayama on cognitive functions and to compare the immediate and training effects of Bhramari pranayama in the study group. An observational study was conducted among 110 students in a medical college aged between 18-22 years for a period of 2 months. The participants were instructed to do 9 rounds of Bhramari Pranayama after which the immediate effect of the Pranayama and 3 weeks of Pranayama practice on cognitive functions was assessed. After training of Bhramari Pranayama for 3 weeks, a statistically significant improvement was observed. The study shows the possibility that Bhramari Pranayama has a beneficial effect on HRV in medical students, as the autonomic balance shifts towards the improvement of parasympathetic tone. The study also showed better attention, concentration and improved cognitive functions immediately after performing Bhramari pranayama which might be due to increased alertness and calmness.

Key Words: Cognitive functions, Pranayama, Stress, Vagal tone.

Introduction

Yoga is an ancient Indian science[1] as well as the way of life which includes the practice of specific posture (Asana) and regulated breathing (Pranayama)[2]. Yoga is a 3000-year-old spiritual and ascetic discipline, which has been designed to bring harmony to the physical, mental, emotional, and spiritual health of an individual[3]. Pranayama is one of the most important yogic practices which can produce different physiological responses in healthy individuals[4]. Pranayama involves manipulation of breath movement[5,6] where the breath is a dynamic bridge between the body and mind. It can be practiced in either slow or fast manner[7]. Pranayama is an art of prolongation and control of breath, which

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helps to bring conscious awareness in breathing, to reshape breathing habits and patterns. Practicing pranayama reduces the effects of stress and strain on various systems by increasing the vagal tone. With the ongoing COVID-19 pandemic, there are heightened feelings of stress, anxiety and depression and pranayama can be an effective way to improve our mental and emotional well-being.

Bhramari pranayama (Humming bee breath) is one of the common slow pranayama practices, which involves inhaling through both nostrils and while exhaling produce sound of humming bee[7]. It could be easily practiced by everyone irrespective of their age or gender. It changes the normal breathing rhythm, with short inhalation and prolonged exhalation producing significant impact in physiological system. Practice of Bhramari Pranayama continuously induces subjective feelings of mind refreshment and blissfulness[7].

Anxiety, stress and mental tensions have almost become inevitable companions of human life especially affecting healthcare course students[6]. Practice of pranayama has found to improve cognitive functions[6]. Cognition is the higher brain function enabling the individual to experience the world by a complex process of interpretation of sensory information. It includes evaluation, categorization and discrimination of stimulus. Executive functions refer to cognitive processes that regulate, control and manage other cognitive processes. Executive functions include working memory, concentration span, scanning and retrieval of stored information and mental flexibility i.e., the ability to shift from on criterion to another in sorting or matching tasks[6].

As there are very few studies on the effects of Bhramari pranayama[2,7] and none compares the immediate and training effect of Bhramari pranayama on cardiovascular and cognitive functions in healthy adolescents, this study has been chosen. The present study aimed to assess the effect of Bhramari pranayama on autonomic functions and cognitive functions and to compare the immediate and training effects of Bhramari pranayama in the study group.

Material and Methods

The observational study was conducted among 110 students in a medical college aged between 18-22 years for a period of 2 months from 30/08/2021 to 30/10/2021.

Data collection method:

The subjects were selected by convenient sampling technique and the selection is based on the following criteria:

Inclusion criteria:

1. Subjects aged between 18-22 years of either gender

Exclusion criteria:

1. Subjects who practiced yoga techniques in past 1 year
2. Subjects with history of previous or current organic diseases (likely to reduce cognition)
3. Subjects who were unable to practice pranayama due to physical abnormalities
4. Subjects with history of chronic respiratory illness
5. Subjects on medication
6. Subjects with history of any acute illness 3 months prior to the study
7. Subjects with history of any surgeries in recent past
8. Athletes, smokers and alcoholic

Brief procedure:

The study was carried out in the department of physiology in our Institution. The approval of the Institutional Research and Ethics committees was obtained prior to the commencement of the study. The subjects were recruited from our institution among the undergraduate medical students. Informed written consent was taken from all the subjects prior to the study. Bhramari Pranayama training was given by a qualified yoga instructor. The participants were instructed to sit in a comfortable posture with spine erect and eyes closed. They were asked to take slow and deep inhalation through both nostrils for 6 seconds, followed by deep and slow exhalation in the same way for 10 seconds with their index finger on both external auditory canal. While exhalation, they were instructed to chant a humming nasal sound like a bee. 9 rounds were done.

The participants were called in groups of 5 to the physiology lab and were instructed to do 9 rounds of Bhramari Pranayama after which the immediate effect of the pranayama on cognitive functions was done. The readings were noted within an interval of 5 minutes. Bhramari Pranayama training was given by the yoga instructor everyday in the evening for 15 minutes for 3 weeks after which training effect of Bhramari Pranayama was taken. A total of 110 subjects with age group (18-22 years) were recruited. The anthropometric measurements (Height and Weight) were recorded and BMI was calculated as: BMI = Weight (Kgs) / Height (Mts)^2.

Cognitive functions:

The following tests will be done to assess the Cognitive functions- Mini mental status examination (MMSE), Wechsler memory scale – revised (WMS-R), Digit letter substitution test (DLST), Digit Symbol Substitution Test (DSST). Scoring was done using the scale.

Mini Mental Status Examination (MMSE): It was done by asking a set of 11 questions under following sections like orientation, registration, attention, calculation, recall and language. The questions are very basic like what is the year, season, date, month etc. The total score was assessed and the participant was categorized as alert/ coma/ stupor/ drowsy.

Wechsler Memory Scale – Revised (WMSR- R): Two different tests were done in this type of cognitive function tests.

(a) Spatial addition subtest: It assess visuo-spatial storage and manipulation in working memory. The participant was shown a grid with blue or red dots on it for 5 seconds. They were asked to remember the location of the blue dots and ignore red dots that appeared on page. The participant was then shown a second page with blue and red dots for 5 seconds, participant then adds the two visual images together. The participant must place the blue dot in the grid in location where they saw blue dots on either pages and white dot in location where blue dots appeared in common.

(b) Design subtest: The participant was shown a page with designs placed in grid. There are 4 times having 4,6,6,8 designs for participant to remember respectively. The participant was asked to remember the designs and the location of designs. After seeing the stimulus page for 10 seconds, the participant was given puzzle grid and cards with designs on them. The participant must select the cards with correct designs and place them in puzzle grid in correct position. After 20-30 minutes of delay, the participant was given the cards to place in the grid. Following the delay recall task, a delayed recognition is administered and scores were calculated for total immediate, immediate content, immediate spatial, total delay, delayed content and delayed spatial.

Digit Letter Substitution Test (DLST): The test was done to assess cognitive function. It consists of one letter - digit pairs (eg. W/1, B/2, T/3, P/4, V/5…J/9) followed by list of alphabets. Under each alphabet, the participant was instructed to write down the corresponding digit within 60 seconds of time period. The correct digit was considered as a score.

Digit Symbol Substitution Test (DSST): The test was done to assess neuropsychological activity of brain. It consists of one digit – symbol pair (eg. 1/-, 7/^, 9/=…) followed by a list of digits. Under each digit the participant should write down the corresponding symbol as fast as possible within 60 seconds. The number of correct symbols within the allowed time is measured and score awarded.

Statistical Analysis

Data were entered in Microsoft - Excel and analysis was done using SPSS version 23. Categorical variable was expressed in frequency and percentage, continuous variables was expressed in mean and standard deviation and paired't' test was used to test the significant difference. p value less than 0.05 was considered to be statistically significant.

Results

Total of 110 MBBS students participated in the study. The demographic characteristics of the study group like age, gender distribution and Body mass index are depicted in Table 1. The age distribution of participants (n=110). Data expressed in frequency (%). About 44% of study participants were belonged to 21 years of age and minimum belonged to 19 years of age with 15%. The gender distribution of participants (n=110). Data expressed in frequency (%). Maximum number study participants were female with 64.5%. The Body Mass Index of the participants (n=110). Data expressed in frequency (%). About 65% of the study participants were in normal weight category according to WHO classification.

Table 1: The cognitive functions on immediate and after the practice of Bhramari Pranayama expressed in mean ± SD

Parameters Measured	Immediate Effect of Bhramari Pranayama	After Bhramari Pranayama Practice	P Value
Mini Mental Status Examination (MMSE)	28.3 ±0.51	27.6 ± 0.53	0.000**
Wechsler Memory Scale-Revised (WMS-R) 1	4.7 ±0.49	4.5 ±0.72	0.045*
Wechsler Memory Scale-Revised (WMS-R) 2	5.6 ±0.60	5.4 ±0.72	0.020*
Digit Letter Substitution Test (DLST)	46.2 ±4.81	43.4 ±5.76	0.000**
Digit Symbol Substitution Test (DSST)	47.9 ±4.76	44.1 ±6.86	0.000**

Comparison of immediate and training values of Bhramari Pranayama on cognitive test parameters. Expressed in mean ± SD. Statistical analysis was done by students' paired 't' test. *p value <0.05, **p value < 0.01.

Table 1 shows the cognitive functions on immediate and after the practice of Bhramari Pranayama expressed in mean ± SD in which there is a statistically significant change in values of MMSE, WMSR 1, WMSR 2, DLST, DSST. $p < 0.05$ was considered to be statistically significant.

Discussion

Yoga is a science that facilitates homeostasis, an ancient way of life intended to improve the quality of life of an individual[8]. Pranayama practice includes voluntary breath regulation, which allows a practicioner to modify physiological functions and mental state within physiological limits[9]. The present study was undertaken to assess the immediate and training effect of Bhramari Pranayama on cognitive functions.

According to the Ancient Yoga Tradition, the breath and the mind are closely interconnected and their influence is bidirectional[10]. The results of our study showed better cognitive functions, immediately after doing the Bhramari Pranayama. This might be because, during pranayama, participants initially focus on breathing at different frequencies of respiration and intend to relax, attention is drawn away from extraneous distracting stimuli. Vagal afferents from peripheral receptors are connected with the nucleus tractus solitarius from which fibers ascend to the thalamus, limbic areas, and anterior cortical areas[3]. During above tidal volume inhalation as seen in Hering Breuer reflex ,stretch of lung tissue produce inhibitory signals in the vagus nerve which ultimately shifts the autonomic nervous system into parasympathetic dominance, that results in a calm and alert state of mind[3,6].

Therefore with continuous pranayama practice, the ability to concentrate will enhance and the changes in mental processing (Eg.focussed attention and reduced stress) are rapidly expressed in the body via the autonomic and neuro endocrine systems. This reorganizes neural representation within the CNS and improve bidirectional communication between the cerebral cortex and the limbic, autonomic, neuroendocrine and behavioural activation[6]. Hence, it is evident that the beneficial psychological effects observed with Pranayama are likely to be a result of both neurohumoral mechanisms, predominantly involving the sympathetic-parasympathetic nervous system[11]. The slight decrease in cognitive function after training may be because of external stress. The immediate effect of bhramari pranayama on cognitive functions was done in a group of 5, but the training effect was done in the classroom due to inevitable reasons which may have lead to distractions.

Conclusion

The present study throws light on how Pranayama is extremely beneficial to mankind in maintaining sound physical and mental health. The findings of the study concludes that, Bhramari pranayama is beneficial for stress reduction. The study also showed better attention, concentration and improved cognitive functions immediately after performing Bhramari pranayama which might be due to increased alertness and calmness. This study was conducted on healthy adolescents, future studies should broaden the current research and should include patients with cardiovascular problems and psychiatric disorders, whose cognitive functions are adversely compromised.

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Conflict of interest: Nil

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Physiological and Motor Performance Characteristics in Rugby and Soccer Players: Data from a Developing Country

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Abstract

This study comparatively presents the physiological, anthropometric, and motor performance characteristics of rugby and football athletes in a developing country. 49 university soccer [(US: N=24, mean age = 22.54±2.24), university rugby (UR: N=25, mean age =21.84±1.81)] and 23 national rugby (NR) (mean age = 26.0±3.33) were recruited. Body weight, body mass index (BMI), waist-to-height ratio (WtHR), hip circumference (HC), waist circumference (WC), waist-to-hip ratio (WHR), blood pressure, heart rate, maximal oxygen consumption (VO_2max), 5-Jump Test (speed), agility, core muscle strength and stability (CMS), muscular endurance (ME), and sit and reach (SRT) were measured. NR significantly had higher age, weight, BMI, WtHR, HC, WC, WHR, SBP, VO_2max, 5JT/BM and ME ($P < 0.05$) compared to others. National and university rugby athletes had better anthropometric and physiological attributes with similar agility performance. To develop rugby in developing country, university soccer athletes could suggest good material, given needed tactical trainings and competitions, for national rugby team athletes.

Key words: Blood pressure, heart rate, maximal oxygen consumption, hip circumference, waist circumference, Speed, agility, core muscle strength and stability, muscular endurance.

Introduction

Rugby and soccer required much running and tackling.[1] Soccer does not involve full-contact as rugby but both engage in physical contact regarded as fair charging for the ball in a distance.[2-4] To attain favorable performance outcomes, rugby and soccer players should possess adequate physical fitness collectively and individual skills.[5,6] Globally, emphasis is placed on the overall physical fitness parameters of rugby players such as muscular strength, muscular endurance, power, agility and flexibility either in a match or training session.[6,7] Considering time and movement patterns, rugby players perform absolutely distinct activities in

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competition related to individual playing position than soccer.[7]

Rugby players are distinguished as backward and forward athletes whereas soccer players categorized as strikers, midfielders, and defenders.[8,9] Based on lower body, rugby players received minimal attention due to collision at the lower extremities relative to soccer.[10,11] Regarding contact sports, soccer players attract less publicity on upper extremity activities than rugby players.[11]

Studies have advocate for well-developed physiological characteristics in elite rugby and soccer games.[7,12-14] Given that physiological profile in rugby and soccer players depend on playing position, there is lower sameness within the playing position in rugby players compared to higher uniformity in soccer players. Also, development of adequate motor performance like muscular strength and endurance, agility, power, and flexibility cannot be overemphasized for success in rugby and soccer competitions.[6,7] Studies have also addressed the combinations of sprinting tests of acceleration, maximal speed, and agility or tests of strength and power, whereas other studies have attempted a more holistic test battery.[7,15-21]

Although rugby and soccer games are two very different sports, but whether the physique and body shape (anthropometric) and motor performance characteristics like power, muscular strength and endurance, agility, and flexibility of athletes in developing countries such as Ghana are different has not been investigated. This study comparatively presents the physiological, anthropometric, and motor performance characteristics of rugby and football athletes in a developing country.

Materials and Methods

Study design: The study adopted a descriptive cross-sectional research design. Participants were drawn from the national rugby (NR) Ghana's first team, a university rugby (UR) and a university soccer (US) team. 72 athletes (US=24, UR=25, and NR=23) were recruited for the study. Participants diagnosed with injury and illness were excluded from the study.

Measurements: Physiological characteristics of heart rate (pulse), systolic and diastolic blood pressure were measured using the Omron blood pressure monitor (HEM-7130 Blood Pressure Monitor). An externally paced running fitness beep test was deployed to used to determine the maximal oxygen consumption (VO_2max) of the participants. The height, body weight, waist circumference, and hip circumference of participants were measured. Body mass index (BMI) value obtained from the body composition analyzer was recorded and analysed in (kg)/ height^{-2} (m^{-2}). The ratio of waist circumference over hip circumference resulted to waist-to-hip ratio were anthropometric characteristics measured. For motor performance, participants performed lower limb explosive power (*5-Jump Test, 5JT)*, Illinois agility, eight stages core muscle strength and stability, isometric wall squat, and Acuflex I Modified Flexibility Sit and Reach Box (Model: 00011) tests.

Statistical analysis: Descriptive statistics of percentiles, mean and standard deviation and inferential statistics analysis of variance (ANOVA). Specific area of differences were analysed with post hoc test of Bonferroni. Significance was set at 0.05 alpha level.

Results

The groups had mean age of 26.0±3.33, 22.54±2.24, and 21.84±1.81 years with 25th percentile of 23.00, 21.00 and 20.00, respectively. For body mass index value, differences were detected among the groups with 95th percentile of 37.720, 34.44 and 25.20. Associating the 95th percentile of BMI to the normative values of World Health Organization (WHO), NR athletes were at risk of obesity II, UR at obesity I, and US at overweight. Table 1 showed the differences in anthropometric, physiologic, and motor performance characteristics of the participants among the groups while table 2 presented post hoc test of Bonferroni results with specific significant difference ($p < 0.05$).

Table 1: Differences in Anthropometric, Physiologic and Motor Performance characteristics of the participants among the Groups (N=72).

Variable	US	UR	NR	Percentile				F	p
	Mean ± SD (N=24)	Mean ± SD (N=23)	Mean ± SD (N=25)	25th	50th	75th	95th		
Age(yrs)	22.54± 2.25	26.00±3.33	21.84±1.82	20.89[a]	22.92	24.82	30.11	18.376	.000*
Height(m)	1.73±6.32	1.77±9.47	1.74±8.59	1.71[a]	1.74	1.79	1.90	1.447	.242
Weight (kg)	67.43±6.59	79.96±13.85	71.96±12.73	65.00[a]	71.25	77.27	101.57	7.139	.002*
BMI (kg/cm^2)	22.35±1.83	26.56±4.87	23.66±4.17	21.50[a]	23.20	25.37	34.20	7.358	.001*
WtHR	0.43±0.03	0.52±0.12	0.47±0.05	0.42[a]	0.45	0.53	0.61	7.968	.001*
HC(cm)	93.15±4.90	83.03±12.28	98.60±19.92	86.05[a]	92.63	98.64	113.80	7.652	.001*
WC(cm)	74.15±3.99	90.80±17.26	84.00±9.80	73.80[a]	78.42	92.50	108.70	12.360	.000*
WHR	0.79±0.03	1.10±0.21	0.82±0.06	0.78[a]	0.82	0.93	1.26	42.580	.000*
SBP(mmHg)	113.21±9.44	130.09±15.05	126.64±13.29	112.75[a]	121.20	132.17	150.73	11.581	.000*
DBP(mmHg)	69.50±7.89	80.00±11.02	86.44±13.89	70.00[a]	77.33	86.00	101.20	14.145	.000*
HR(bpm)	59.79±7.74	66.74±12.59	75.92±12.36	58.25[a]	65.86	75.20	92.90	12.983	.000*
VO_2max (ml/kg/min)	43.91±6.18	46.41±5.14	35.24±6.53	35.82[a]	41.91	47.72	52.45	23.218	.000*
5JT/BM (cm/kg)	3.02±0.62	3.83±0.85	3.73±1.68	2.82[a]	3.37	3.91	4.99	3.410	.039*
Agility (sec)	12.37±0.81	12.52±1.02	12.78±2.25	12.03 [a]	12.69	13.38	14.19	.458	.634
CMS (sec)	7.21±1.52	7.57±0.79	6.88±1.20	6.50[a]	7.52	0.00	0.00	1.898	.158
ME(sec)	42.48±10.96	53.52±11.72	37.84±14.08	35.00[a]	41.57	52.17	68.93	10.071	.000*
SRT(cm)	34.75±7.21	32.57±9.12	33.56±7.35	28.43[a]	33.67	38.83	47.60	.449	.640

Data reported in Mean value and Standard deviation (Mean± SD); [a] Percentiles are calculated from grouped data. Body weight, body mass index (BMI), waist-to-height ratio (WtHR), hip circumference (HC), waist circumference (WC), waist-to-hip ratio (WHR), blood pressure, heart rate, maximal oxygen consumption (VO2max), 5-Jump Test (5JT/BM), agility, core muscle strength and stability (CMS), muscular endurance (ME), and sit and reach (SRT) were measured. *p<0.05, Mean value and Standard deviation: Mean± SD, SS: Sum of Square, MS: Mean Square, CI: Confidence Interval, MD= mean Difference

Table 2: Post hoc tests of Bonferroni

Variable	(I)	(J)	Mean Diff.(I-J)	P	95% CI
Age (yrs)	NR	US	3.458*	0.000	1.65, 5.26
		UR	4.160*	0.000	2.37, 5.95
Weight(kg)	NR	US	12.527*	0.001	4.30, 20.75
BMI (kg/cm^2)	NR	US	4.202*	0.001	1.46, 6.95
		UR	2.901*	0.033	0.18, 5.62
WtHR	NR	US	0.093*	0.000	0.04, 0.15
HC (cm)	US	NR	10.124*	0.046	0.15, 20.10
	UR	NR	15.574*	0.001	5.70, 25.45
WC (cm)	NR	US	16.659*	0.000	8.38, 24.94
	UR	US	9.854*	0.012	1.74, 17.96
WHR	NR	US	0.307*	0.000	0.22, 0.40
		UR	0.286*	0.000	0.20, 0.38
SBP (mmHg)	NR	US	16.879*	0.000	7.73, 26.03
	UR	US	13.432*	0.001	4.47, 22.39
DBP(mmHg)	NR	US	10.500*	0.006	2.46, 18.54
	UR	US	16.940*	0.000	9.07, 24.81

Variable	(I)	(J)	Mean Diff.(I-J)	P	95% CI
HR (bpm)	UR	US	16.128*	0.000	8.33, 23.93
		NR	9.181*	0.017	1.30, 17.06
VO_2max(ml/kg/min)	US	UR	8.671*	0.000	4.46, 12.88
	UR	UR	11.173*	0.000	6.92, 15.43
ME (sec)	NR	US	11.043*	0.009	2.19, 19.89
		UR	15.682*	0.000	6.92, 24.45

**p<0.05, CI: Confidence Interval*

Discussion

This study investigated the physiological, anthropometric, and motor performance characteristics of rugby and football athletes in a developing country. Results showed significant difference in weight, body mass index, waist to height ratio, waist to hip ratio, blood pressure, heart rate and VO_2max, and muscular endurance among the categories differences which support earlier submissions that variations in anatomical body shapes may influence physical performance.[22-24]

Although there were significant differences in physiological and motor performance among the groups, similarities showed in some characteristics. For example, players' height and weight in the NR team were more than that of UR and US teams. Differences in professional recruitment strategy where specific standard criteria should be met to qualify for selection could be implicated.[16] Roscoe established that NR players had higher, broader, and stability body shape than UR and US teams.[24] This suggests that university athletes have no strict recruitment process comparatively since players are drawn from the pool of students admitted. Hence, university sports coaches, irrespective of sports discipline, would expectedly work vigorous with the materials within reach to attain optimal success.

Distinctions were also discovered in body mass index (BMI) which correlates with the body mass of US players.[24] US and UR players had better waist to hip ratio and low health risk compared to UR and NR players with high health risk.[25-28] Study reiterates that in contact situation extra body fat function as a supportive cushion especially among the forwards, though directly affect players' power-to-weight ratio, reduces boost ability and increase energy consumption.[29]

Among the groups, US players had normal blood pressure and lowest heart rate while UR and NR players were prehypertensive and hypertensive respectively. This might be related to changes in the product of demand in respiratory functioning.[30] Low heart rate at rest implies better breathing capacity when blood is elevated, reduced stress level, drug impact and inadequate blood flow to the athletes' subconscious within the game.[31] The ability to understand playing patterns and perform efficient actions amid pressure and stress was an element of successful athletes. This study showed that NR players had dominant VO_2max over US and UR players accordingly. The rational, considering NR, US, and UR at large, will give credence to the levels of participation. It can also be seen that UR was emerging game compared to US games in Ghana. Elite athletes need agility levels to withstand the physiological constraints and execute the variety of offensive and defensive capabilities that competition needed.[32-33]

Conclusion

There are differences and similarities in the physiological, anthropometric, and motor performance characteristics of rugby and football athletes in a developing country. University soccer athletes could be good material, given needed tactical trainings and competitions, for national rugby team athletes. Considering the physiological characteristics, electrocardiogram assessment will certainly add strength our findings.

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A Study of Pattern and Prevalence of Cardiovascular Autonomic Neuropathy(CAN) in Type 2 Diabetes Mellitus Patients

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Abstract

Background: Cardiovascular autonomic neuropathy(CAN) is a common underdiagnosed complication in patients with type -2 diabetes mellitus in clinical practice. CAN is associated with increased morbidity and mortality among diabetics. Only few studies are available in literature about pattern and prevalence of CAN among type-2 diabetics in developing countries like India where burden of the disease is high.

Aim : To study the pattern and prevalence of CAN in type 2 Diabetes mellitus patients and to study the sympathetic and parasympathetic abnormalities related to cardiovascular system in patients with type 2 Diabetes mellitus.

Materials and Methods: The study design was observational Retrospective and was conducted at Department of Physiology, Thanjavur Medical college, between april to june 2022. We recruited 80 patients (55 male and 25 female) with type 2 Diabetes mellitus of age 30 years and above from Medicine and Non-Communicable Diseases (NCD) Outpatient Department of Thanjavur Medical college hospital. The study was approved by Institutional Ethical committee(IEC). After getting informed written consent from all the participants, age, duration of diabetes, systolic blood pressure and HbA1c levels were recorded. Patients were made to undergo Autonomic Function Tests(AFT) in Research Laboratory. Assessment of CAN was done retrospectively and scoring was done using three different scoring system namely Ewing's, Bellavere's and AIIMS criterias.

Statistical Analysis and Results: Mean age was 48.86(±5.694) years and duration of diabetes was 108.48(±25.039) months. Mean systolic blood pressure(SBP) was 132.10(±25.039)mmHg and HbA1c level was 8.63(±0.46368). Statistical Analysis was done using unpaired 'T' and Chi-Square tests. The abnormality of single autonomic test ranged from 6% in valsalva ratio to 46.25% in cold pressor test. CAN was found in 14 and 20 patients by Ewings's and Bellavere's criteria respectively. AIIMS criteria revealed 43 patients with isolated sympathetic abnormality and 37with parasympathetic abnormality and 25 with both abnormalities. Duration of diabetes, systolic blood pressure(SBP) and and HbA1c levels were not significantly correlated with autonomic abnormality in our study. Only Valasava Ratio(VR) correlates with autonomic abnormality significantly(P value<0.05).

Conclusion: CAN is a common microvascular abnormality in type 2 diabetes mellitus. We have to highlight the importance of regular testing of autonomic functions in type 2 diabetes mellitus to prevent mortality due to cardiovascular complications.

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Key words: Cardiovascular autonomic neuropathy(CAN), Autonomic function Test(AFT), HbA1c, Valsalva ratio(VR)

Introduction

Dysfunction of the Autonomous Nervous system is an underdiagnosed cause of excessive morbidity and mortality in adults with diabetes. Among Non-Communicable Diseases, Type 2 Diabetes Mellitus(DM) is emerging as a great threat and burden to Indian population as India is becoming world diabetes capital[1].

Complications of Diabetic Autonomic Neuropathy are often missed as symptoms are mild and not life threatening. CAN is the major underdiagnosed complication in type -2 DM as its prevalence is 31-73% and its annual incidence is 2%[1].

Early identification of CAN in type -2 DM patients can lead to early intensive interventions targeting life style, glycemic control and risk factors that can slow the progression of CAN[1]. Common symptoms of CAN which are often missed and under diagnosed are exercise intolerance, resting tachycardia, heat intolerance, arrhythmias, dry skin and orthostatic hypotension[2].

CAN in type -2 DM is the result of complex interactions among degree of glycemic control, disease duration, age-related neuronal attrition and systolic and diastolic blood pressure[3].

Materials and Methods

This study was an observational retrospective study, conducted at Department of Physiology, Thanjavur medical college. The study was approved by IEC- 961/2022. Written informed consent were obtained from all study participants. We have selected 80 patients of type 2 DM ofboth sexes of age 30 yrs and above attending Medicine and NCD Outpatient Department. 55 males and 25 females participated in the study. This study was conducted from april to june 2022. Exclusion Criteria were Normal healthy adults, Type 2 diabetics who were newly diagnosed and those with disease less than one year, previous history of coronary artery disease, hepatic, respiratory, renal, cerebrovascular, thyroid or other endocrine abnormalities, previous history of ECG abnormalities and with any history of acute illness within past 48 hrs. Female patients during menstrual cycles were excluded. Patients should not have consumed beverages containing caffeine or alcohol within past 12 hrs and patient should not be on drugs like betablockers or sympathomimetics or vasodilators or diuretics or antiarrhythmics or calcium channel blockers. Patients engaged in strenuous exercise in the preceding 24 hrs were excluded from the study.

Detailed history including treatment, dietary, lifestyle and family histories and duration of DM were recorded. Baseline investigations including SBP in mmHg using mercury Sphygmomanometer was recorded and about 3ml of blood samples were collected from the antecubital vein for HbA1c. HbA1c was analysed using automatic analyser (A1c 2.2 Glycohemoglobin Analyser HLC-723GHb). ECG recording was done using ECG machine (BPL Cardiart 6208 View 3 Channel).

Cardiovascular Autonomic Function tests:

The six autonomic function tests were done for all the patients as per standard protocol[4]. All the tests were done after proper instructions.

1. Deep breathing test: The patient was asked to lie down in the supine position. Lead II ECG was recorded for heart rate and respiration monitoring was done for 30 seconds.The subject was asked to breathe deeply at a rate of six breaths per minute(allowing 5 seconds each for inspiration and expiration). Maximum and minimum heart rate with each respiratory cycle were recorded and average was taken.

2. Valsalva Maneuver:The patient was made to sit comfortably and ECG electrodes were connected and recording was done. The patient was asked to blow or exhale forcefully into the mouthpiece connected to the sphygmomanometer after closing the nostrils with nose clip and was asked to maintain the expiratory pressure at 40mmHg for 10-15 seconds. Recording of ECG was done throughout and 30 seconds before and after the procedure. After 30 seconds,the patient was asked to release the pressure. Valsalva Ratio was calculated from the longest RR interval during phase IV and shortest RR interval during phase II.

3. Handgrip test: Baseline BP was recorded and the patient was asked to press handgrip dynamometer at 30 percent of maximum contraction for 4 minutes. The DBP was recorded at first, second, third and fourth minute of contraction. The rise in the DBP above baseline was recorded.

4. Cold Pressor test: Baseline BP was recorded. The patient was asked to immerse the right hand into the container containing cold water(at or below 4 degree Celsius) for 60 seconds. The DBP was recorded at 30 and 60 seconds. The rise in the DBP above baseline was recorded.

5. Lying to standing test: the patient was asked to lie down in supine position. The basal BP and ECG were recorded. Then the patient was asked to stand immediately within 3 seconds from supine position. ECG was recorded continuously during the procedure at 0.5th,1st,2nd,2.5th and 5th minute.30:15 ratio was calculated from ECG (i.e) ratio of RR interval at 15th beat and RR interval at 30th beat. Record blood pressure at 2nd and 5thminute. The fall in SBP was recorded.

6. Head Up Tilt test: After proper instructions, the patient was asked to lie down in Head –Up tilttable for 5 minutes. Baseline BP and ECG were recorded. Then the patient was made to stand erect 80 degrees passively from supine position within 15 seconds and was maintained in that position for 5 minutes. Immediately heart rate and BP were recorded for 5 minutes. The maximum fall in SBP was recorded.

1. Categorization of patients as per Bellavere criteria[5]

1. Deep breathing test(delta heart rate)

2. Valsalva maneuver(Valsalva Ratio)

3. Lying to standing(30:15 ratio)

The scores were added and CAN was categorized as follows

0-1 = no CAN

2-3 = early CAN

4-6 = definite CAN

2. Categorization of patients as per Ewing´s criteria[6]

All the tests were included except cold pressor test.

The scores were added and CAN was categorized as follows

Normal = all tests normal or one test borderline

Early = one of the three heart rate tests abnormal or two borderline

Definite = two heart rate tests abnormal

Severe = two heart rate tests abnormal + one or both BP tests abnormal

3. Categorization as per AIIMS criteria[2]

Test for parasympathetic parameters

1. Deep breathing test (delta HR)

2. Valsalva maneuver (VR)

3. Lying to standing (30:15 ratio)

Tests for sympathetic parameters

1. Handgrip test (change in DBP)

2. Cold pressor test (change in DBP)

3. Lying to standing test (change in SBP)

Table 1: Test for Assessment of Autonomic Function

Test	Parameter	Criteria	Category	Score
Deep Breathing test	Delta HR(bpm)	>15 11-14 <10	Normal Borderline Abnormal	0 1 2
Valsalva Maneuver	VR	>1.21 1.11-1.20 <1.10	Normal Borderline Abnormal	0 1 2

Test	Parameter	Criteria	Category	Score
Handgrip test	Change in DBP(mmHg)	>16 11-15 <10	Normal Borderline Abnormal	0 1 2
Cold pressor test	Change in DBP(mmHg)	<10 11-20 >20	Normal Borderline Abnormal	0 1 2
Head-Up tilt	Fall in SBP(mmHg)	<10 11-20 >20	Normal Borderline Abnormal	0 1 2
Lying to standing test	30:15 ratio	>1.04 1.01-1.03 <1.01	Normal Borderline Abnormal	0 1 2

Statistical Analysis

Statistical analysis was done using the SPSS statistics software version 20. All the data were shown as the mean or as percentages. Continuous variables were analysed by Unpaired student´s t-test. Categorical variables were compared by the Chi-square test. P value <0.05 was considered as statistically significant.

Results and Discussion

The mean age of 80 patients who participated in the study was 48.86 ± 5.694 years. Among them, 55 were male and 25 were female which corresponds to 68.8% and 31.3% respectively. Mean duration of diabetes was 108.48 ± 25.039 months. Mean HbA1c value was 8.63 ± 0.46368.Mean systolic blood pressure was 132.10 ± 6.689 mmHg. Out of 80 patients, only 49 patients were able to do all autonomic function tests. All patients were not able to do physical effort related tests like Valsalva Maneuver, Hand grip test and deep breathing test .Valsalva maneuver was not done in patients with diabetic retinopathy. Table II shows results of individual test done in 80 patients. Table III shows the scoring based on Bellavere, Ewing´s and AIIMS criteria respectively. CAN was found in 40,29 and 43 patients according to Bellavere, Ewing's and AIIMS criteria respectively.

Figure I shows bar diagram of genderwise loss of autonomic function in diabetes which shows no specific variation among tests between genders except valsalva maneuver. Table IV shows statistical inference of continuous variable using Unpaired ´t´ test which shows age, duration of diabetes and HbA1c were not significant. Table V shows statistical inference of categorical variable using Chi-square test and correlation coefficient and Pearson's correlation analysis between independent variable.

Table 2: Results of Autonomic Function Test Done in The Patients

Test	Parameter	Total	Borderline	Abnormal	%Abnormal
Deep Breathing test	Delta heart Rate(bpm)	80	28	30	35
Valsalva Maneuver	Valsalva Ratio	50	3	5	6
Handgrip test	Change in diastolic blood pressure(mmHg)	70	32	21	45.71
Cold pressor test	Change in diastolic blood pressure(mmHg)	80	37	17	46.25

Test	Parameter	Total	Borderline	Abnormal	%Abnormal
Head up tilt	Fall in systolic blood pressure(mmHg)	80	10	28	12.5
Lying to standing test	30:15 ratio	80	14	1	17.5

Table 3: Categorization as Per Bellavare,Ewing's and Aiims Criteria

BELLAVRE CRITERIA	NO OF PATIENTS	
NORMAL(SCORE 0-1)	40	
EARLY(SCORE 2-3)	20	
DEFINITE(SCORE 4-6)	20	
EWING'S CRITERIA	**NO OF PATIENTS**	
NORMAL	51	
EARLY	15	
DEFINITE	–	
SEVERE	14	
AIIMS CRITERIA	**BORDERLINE**	**ABNORMAL**
PARASYMPATHETIC(IRRESPECTIVE OF SYMPATHETIC STATUS)	29	29
PARASYMPATHETIC BUT NO SYMPATHETIC ABNORMALITY	17	1
SYMPATHETIC(IRRESPECTIVE OF PARASYMPATHETIC STATUS)	17	42
SYMPATHETIC BUT NO PARASYMPATHETIC ABNORMALITY)	17	14
BOTH	10	25

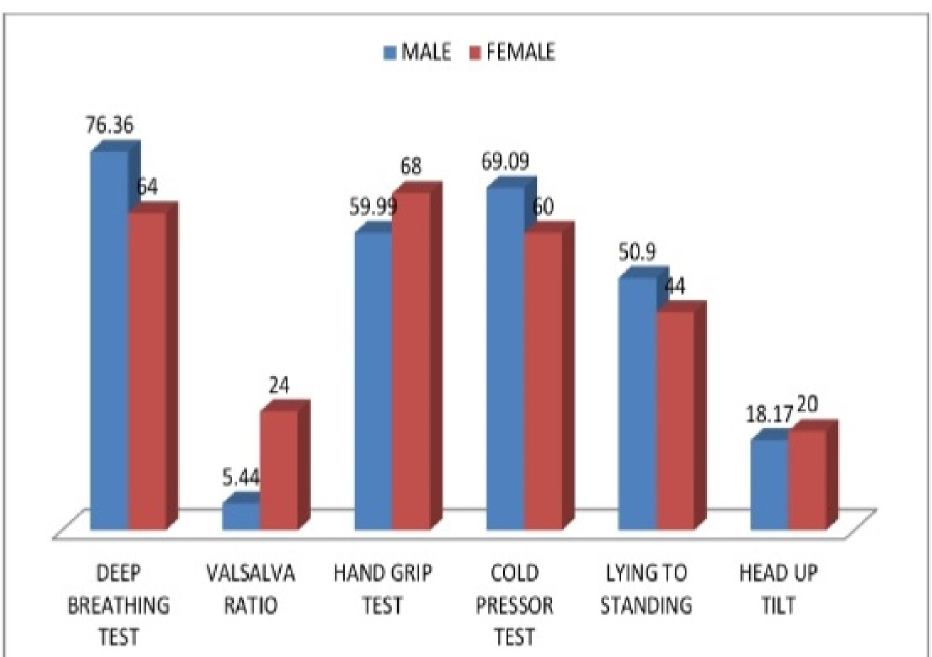

Figure 1 : Genderwise Loss of Autonomic Function in Diabetics

TABLE 4: STATISTICAL INFERENCE BY UNPAIRED´ t´ TEST

SEX	N	Mean	S.D	Statistical inference
SYSTOLIC BP(mmHg)				
Male	55	131.56	6.321	T=1.065 Df=78 0.290>0.05 Not Significant
Female	25	133.28	7.436	
HbA1C %				
Male	55	8.6418	.47442	T=0.158 Df=78 0.875>0.05 Not Significant
Female	25	8.6240	.44840	
DURATION OF DIABETES(months)				
Male	55	109.38	24.422	T=0.478 Df=78 0.634>0.05 Not Significant
Female	25	106.48	26.754	

TABLE 5: STATISTICAL INFERENCE BY CHI-SQUARE TEST AND CORELATION COEFFICIENT

	SEX						Statistical inference
	Male		Female		Total		
	n	%	n	%	n	%	
DEEP BREATHING TEST-							
Abnormal	19	34.5%	9	36.0%	28	35.0%	X^2=0.571 Df=2 0.751>0.05 Not Significant
Borderline	22	40.0%	8	32.0%	30	37.5%	
Normal	14	25.5%	8	32.0%	22	27.5%	
VALSALVA MANEUVER							
Not able to do test	23	41.8%	7	28.0%	30	37.5%	X^2=8.298 Df=3 0.040<0.05 Significant
Abnormal	1	1.8%	2	8.0%	3	3.8%	
Borderline	1	1.8%	4	16.0%	5	6.3%	
Normal	30	54.5%	12	48.0%	42	52.5%	
HAND GRIP TEST -							
Not able to do test	8	14.5%	3	12.0%	11	13.8%	X^2=0.979 Df=3 0.806>0.05 Not Significant
Abnormal	20	36.4%	12	48.0%	32	40.0%	
Borderline	14	25.5%	5	20.0%	19	23.8%	
Normal	13	23.6%	5	20.0%	18	22.5%	
COLD PRESSOR TEST -							
Abnormal	27	49.1%	12	48.0%	39	48.8%	X^2=1.054 Df=2 0.590>0.05 Not Significant
Borderline	11	20.0%	3	12.0%	14	17.5%	
Normal	17	30.9%	10	40.0%	27	33.8%	
HEAD UP TILT TEST -							
Abnormal	7	12.7%	3	12.0%	10	12.5%	X^2=0.349 Df=2 0.840>0.05 Not Significant
Borderline	21	38.2%	8	32.0%	29	36.3%	
Normal	27	49.1%	14	56.0%	41	51.3%	
LYING TO STANDING 30:15							
Abnormal	9	16.4%	5	20.0%	14	17.5%	X^2=0.591 Df=2 0.744>0.05 Not Significant
Borderline	1	1.8%	0	.0%	1	1.3%	
Normal	45	81.8%	20	80.0%	65	81.3%	
Total	55	100.0%	25	100.0%	80	100.0%	

STATISTICAL ANALYSIS BY CORRELATION COEFFICIENT							
INDEPENDENT VARIABLE	**n**	**Mean**	**S.D**		**Age (yrs)**	**Systolic BP (mmHg)**	**Duration of Diabetes (months)**
Age (yrs)	80	48.86	5.694	r	—	.094	.126
				sig.	—	.407	.264
Systolic BP (mmHg)	80	132.10	6.689	r	.094	—	.097
				sig.	.407	—	.393
Duration of Diabetes (months)	80	108.48	25.039	r	.126	.097	—
				sig.	.264	.393	—

Our study reveals sympathetic abnormality was more common than parasympathetic component and abnormality in a single test which varies from 6% in VR to 46.25% in cold pressor test. The study shows only VR correlates significantly with CAN as p value<0.05. The autonomic score does not significantly correlates with age of the patient and duration of diabetes as well as HbA1c level. The age of the patient at which diabetes is diagnosed is variable.

CAN is a serious complication of diabetes that is often associated with poor prognosis and can result in severe postural hypotension, exercise intolerance, enhanced intraoperative instability and increased incidence of myocardial infarction or ischemia and may even predict the development of stroke[7]. According to a meta-analysis, the overall mortality rates over periods up to 10 years were 30.4% for people with diabetes and CAN detected by heart rate variability (HRV) compared with 13.4% without CAN[7].

CAN is the most life threatening complication which results from damage to the autonomic nerves that supply the heart, causing abnormal heart rates and rhythms. The hallmark and earliest indicator of subclinical and symptomatic cardiac autonomic dysfunction is reduced Heart Rate Variability(HRV)[7].

Postural hypotension is the clinical hallmark of CAN in diabetics and is characterized by weakness, faintness, dizziness, visual impairment and even syncope following the change in position from lying to standing[7]. A meta-analysis study revealed an increased risk of silent myocardial ischemia during exercise by 1.96% in people with diabetes and CAN compared with those without CAN[7].

Parasympathetic neuropathy in type -2 diabetics is associated with features of the insulin resistance syndrome[8].

Conclusion

AFTs are simple and cost effective which can be done in routine outpatient services. The purpose of this study is to highlight the importance of testing of autonomic functions during regular outpatient followup in long standing type 2 diabetics. An improvement in autonomic balance by recognition of autonomic imbalance in type 2 diabetics is vital to reduce cardiovascular events and early mortality. Early symptoms and signs of autonomic dysfunction like Valsalva maneuver, resting heart rate, BP responses to standing should be elicited from all type 2 diabetics attending OPD irrespective of duration of disease. Early detection and intervention of CAN is necessary in long standing type 2 diabetics so that rigorous regime including lifestyle modification, physical activity and judicious use of medicines for strict glycemic control can be implemented.

Introduction of new therapeutic agents in treatment regime to improve autonomic functions in type 2 diabetics should be considered. Our participants were motivated to adopt regular treatment and to achieve strict glycemic control and to advocate healthy life style modifications.

Limitations of the Study:

The study was retrospective and was limited to 80 patients. The duration of study period was also very short. The future studies are planned with larger population and with more study duration so that we can able to correlate significantly the duration of diabetes with the autonomic abnormality.

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