10.5005/jp-journals-10036-1195

ORIGINAL ARTICLE

Study of Variability in Heart Rate, Peripheral Blood Flow and Pulse Wave Morphology Index in Health and Disease

[1]Jayaprakash Alva, [2]Shrinath Rao, [3]Boodyar S Rai, [4]Vivek Sakthidharan, [5]Pomaleri K Swapna, [6]Rajesh K Jain, [7]Sushma N Bhat, [8]Manasi S Sawant, [9]Ghansyam D Jindal

ABSTRACT

Importance of heart rate variability (HRV) came into existence with the observations of Hon and Lee in 1965 and had been used ever since for objective assessment of automatic nervous system (ANS) as well as in intensive care monitoring. Blood pressure (BP) variability has been investigated less extensively because of invasive nature or high cost of beat-to-beat BP measurement. Peripheral blood flow (PBF) variability and morphology index variability (of the peripheral pulse) have also been sparingly studied because of the paucity of physiological and clinical correlation. In continuation of these studies, the peripheral pulse from the right wrist has been recorded in 207 control subjects and 207 patients (suffering from different diseases) in resting condition (supine), and HRV, blood flow variability and morphology index variability has been derived from this data. To take care of large scatter in the value of variability parameters in control subjects, multiple observations have been made on the subject, and the average value of variability parameter has been considered for analysis in conformity with central limit theorem (CLT). The control data has shown a remarkable decrease in standard deviation and coefficient of variation and enabled use of parametric statistical analysis. Comparison of variability parameters in health and disease has been observed to be consistent with the nonparametric analysis namely Mann–Whitney test , with a correlation coefficient of 0.8675. Based on this analysis, general and specific parameters, which change in the presence of a disease, have been identified for different diseases. The observations are reported here.

Keywords: Blood flow variability, Central limit theorem (CLT), Coefficient of variation, Heart rate variability (HRV), Morphology index variability.

How to cite this article: Alva J, Rao S, Rai BS, Sakthidharan V, Swapna PK, Jain RK, Bhat SN, Sawant MS, Jindal GD. Study of Variability in Heart Rate, Peripheral Blood Flow and Pulse Wave Morphology Index in Health and Disease. MGM J Med Sci 2018;5(3):101-108.

Source of support: Board of Research in Nuclear Sciences, Department of Atomic Energy, Government of India.

Conflict of interest: None

[1]Dean, [2]Professor and Head, [3,9]Professor, [4,5]Assistant Professor, [6]Scientific Officer (G), [7]Senior Research Fellow, [8]Scholar

[1,3]Father Muller Medical College, Kankanady, Mangaluru, Karnataka, India

[2]Department of Materia Medica, Father Muller Homeopathic Medical College, Deralakatte, Mangaluru, Karnataka, India

[4]Department of Homeopathic Pharmacy, Father Muller Homeopathic Medical College, Deralakatte, Mangaluru, Karnataka, India

[5]Department of Medicine, Father Muller Medical College, Kankanady, Mangaluru, Karnataka, India

[6]Electronics Division, Bhabha Atomic Research Centre, Trombay, Mumbai, Maharashtra, India

[7]Board of Research in Nuclear Sciences, Electronics Division, Bhabha Atomic Research Centre, Trombay, Mumbai, Maharashtra, India

[8]Department of Bio-Technology, MGM Institute of Health Sciences, Kamothe, Navi-Mumbai, Maharashtra, India

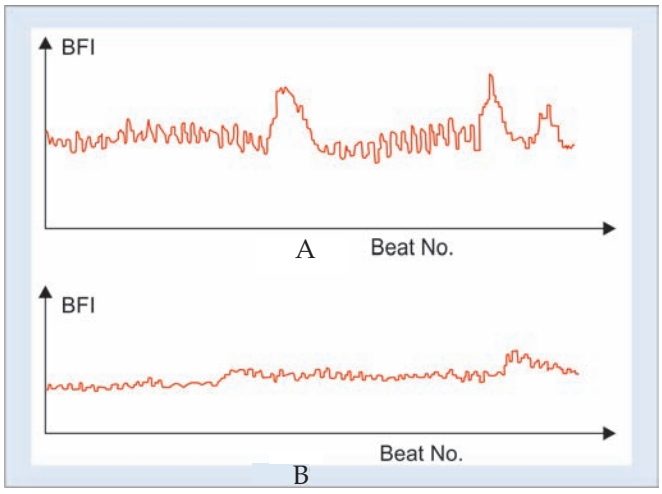
[9]Bio-Medical Engineering Department, MGM College of Engineering andTechnology, Kamothe, Navi Mumbai, Maharashtra, India

Corresponding Author: Ghansyam D Jindal, Professor, Bio-Medical Engineering Department, MGM College of Engineering and Technology, Kamothe, Navi Mumbai, Maharashtra, India, Mob:+919322275221, e-mail: gd.jindal@gmail.com

INTRODUCTION

Variability is the sign of life. In a living being rigidity like that of a nonliving material is considered a sign of poor health. Changes in physiological parameters brought by internal mechanisms in body at rest are termed as physiological variability (PV). The history of PV folds back to 1965, when Hon and Lee observed diminution of beat to beat variation in fetal heart rate during labor fetal distress.[1] Variability can be studied with the help of measurable physiological parameters such as heart rate, systolic, diastolic and mean BP, PBF, body temperature; respiration, electrical activity of the brain, motility of intestines, secretion of endocrine glands and so on. Heart rate (HR), BP and PBF are the most studied variations[2] because of the ease of their recording.

A healthy person has an average heart rate of around 72 beats per minute. It is however never constant as it changes continuously depending upon the physical and mental activity of the subject. Study of these fluctuations or variability, even when the subject is physically inactive is of immense interest to scientists and physiologists. An enormous volume of research has gone into HRV around the globe and resulting in its use in the objective assessment of the ANS and several clinical conditions. Limited amount of work has been done on BP variability because of the use of invasive catheter for BP monitoring or prohibitive cost of noninvasive beat to beat BP monitors. PBF recorded for long intervals in controls and hypertensive subjects has shown prominent and suppressed rhythmic variations in controls and patients respectively,[2] as shown in Figure 1.



Figs 1A and B: Blood flow index plotted against beat number in (A) control subject and; (B) a hypertensive subject. Suppressed variation of the low frequency as well as high frequency rhythm is observed in (B). High frequency rhythm is synchronous with respiration

Based on a large number of studies carried out on short-term PV, three rhythms, commonly known as very low frequency (VLF), low frequency (LF) and high frequency (HF), have been identified for variability analysis, in the frequency ranges 0.003 to 0.04, 0.041 to 0.15 and 0.151 to 0.40 Hz respectively. Ultra low frequency (ULF) rhythm is considered up to 0.003 Hz and is analyzed for 24-hour data. Akselrod et al.[3] has measured HRV in trained, healthy, conscious, un-anesthetized dogs following parasympathetic block with the help of glycol pyrrolate (0.01 mg/kg intravenous) and sympathetic block with the help of propranolol (0.1 mg/kg intravenous). They have observed that parasympathetic block drastically reduces the amplitude of HF peak and LF peak and combined sympathetic and parasympathetic block abolishes all heart rate variation resulting in a metronome-like heartbeat. Many studies[3-5] have gone into the physiological origin of these peaks; the general consensus is that HF peak represents vagal activity and LF peak is contributed by sympathetic as well as a parasympathetic activity; VLF is still more complex and may partly relate to baroreceptor reflex and renin angiotensin system.

Physiological correlation mentioned above has led to a large number of investigations in the field of PV.[6,7] Pagani et al. and Bianchi et al. have shown nearly 20 fold decrease in the total power and HF power in patients with diabetic neuropathy.[8,9] They have shown that HRV analysis can detect diabetic neuropathy in the early stages before the symptoms appear. Copie et al. have shown HRV as a remarkable tool for assessment of the therapeutic response of medicines.[10] They have shown the effect of bisoprolol in patients with cardiac failure; administration of placebo fails to produce any significant change in the HF power, whereas that of bisoprolol causes 50 to 80% increase. Another study has reported HRV analysis in human cardiac transplant recipients.[11] It has been observed that in such cases restoration of HF peak is suggestive of innervations of the transplanted heart. A large number of studies have been conducted on hypertensive subjects. It has been observed that systolic, diastolic and mean BP correlate positively with LF power and the ratio of LF power and HF power.[12] Diastolic BP correlates negatively with HF and total power. In general, increased LF power and decreased HF power are associated with hypertension.[13]

Studies on HRV, PBF variability (PBFV), morphology index variability (MIV) and stroke volume variability (SVV) conducted in the past,[14-17] on patients suffering from different diseases, have shown general changes in variability parameters as shown in Tables 1 and 2. All these studies have not led to the specific manifestation of different diseases on variability spectra (HRV, PBFV, and MIV); probably be because of the large coefficient of variation of variability parameters in controls (60 to 100%). In a recent study, application of CLT has lowered the coefficient of variation and helped in identifying unique changes in harmonic analysis.[18] In view of the above, repeat investigations have been carried out with a modified protocol. Accordingly, PV has been studied in control subjects and patients with ischemic heart disease (IHD), systemic hypertension (HT), pulmonary tuberculosis (TB), cirrhosis of liver (CoL), lung cancer (LC), stomach cancer (SC) and acquired immune deficiency syndrome (AIDS). Results are briefly described in the following sections.

MATERIALS AND METHODS

Control subjects (207) were subjected to this investigation at Father Muller Homeopathic Medical College, Mangaluru under a research project sponsored by Board of Research in Nuclear Sciences (BRNS), Department

Table 1: Behavior of physiological variability in different diseases

Sr. No.	Disease	No. of subjects	Increase in total power	Increase in VLF		Increase in LF		Decrease in HF	
				Area	Amplitude	Area	Amplitude	Area	Amplitude
1.	Diabetes	9	--	HRV, SVV	HRV, SVV, PBFV	–	PBFV	PBFV	HRV, PBFV
2.	Hypertension	31	HRV, SVV	PBFV	HRV, SVV, PBFV	–	–	–	–
3.	Cirrhosis	16	HRV	–	–		PBFV	HRV	–
4.	Tuberculosis	6	–	–	–	HRV, SVV	SVV	–	–
5.	AIDS	19	–	HRV	–	–	PBFV	HRV,	HRV, PBFV
6.	Sterility	5	HRV	SVV	PBFV, SVV	PBFVR	PBFV	–	–
7.	Hypothyroid	23	–	HRV,	HRV, PBFV	–	–	–	HRV

of Atomic Energy (DAE), Government of India (GoI). These subjects were taken from the staff members and students of the college. A patient group comprising 46 with TB, 23 with HT, 31 with IHD, 31 with AIDS, 21 with CoL, 20 with SC and 35 with LC were recorded at Father Muller Medical College, Mangalore under another research project sponsored by BRNS, DAE. These subjects were derived from the outpatient departments of the institution. Age and sex profile in these subjects is given in Table 3. Both the studies were given clearance by respective institution's ethics committee.

The recordings were taken with the help of an instrument known as peripheral pulse analyzer (PPA), a research product of Electronic Division, Bhabha Atomic Research Centre (BARC), and Mumbai. The recordings were taken by considering the guidelines and protocol designed for the studies on HRV.[5,19] The instrument records impedance plethysmogram (rate of change of electrical impedance as a function of time "dZ/dt") at the wrist level with the subject in supine for 275 seconds. Twenty-five recordings in each of the control subjects and five recordings in each of the patients have been taken around the same time of the day, i.e., 2 hours after breakfast. In control subject the readings were taken on three consecutive days considering the convenience of the subjects. The subject was made to lie down on a couch and relax for a period of 10 minutes. With the subject continuing to be in supine, carrier electrodes C1 and C2 were applied around the elbow and palm in the right upper extremity in the form of loop around the extremity segment. Sensing electrodes S1 and S2 were applied on the proximal and distal part of the wrist. Clicking on the "acquire" button in the application software starts data acquisition. Acquisition stops automatically after 275 seconds. In case of motion artifact, acquisition can be stopped any time and a fresh acquisition can be done. After 275 seconds, data is saved in a file with subject identification. Data is re-acquired from the same subject for multiple readings.

The processing software used for processing this data is also from the electronic division, BARC, Mumbai, known as physiological variability analyzer. A click on this application software displays a graphic user interface (GUI) panel on the screen. A click on load button on the panel allows loading a pre-acquired data file. After selecting the signal to be processed and variability to be obtained, one can go to MARK panel. Figure 2 shows the GUI of the MARK panel. In this panel, the acquired signal is displayed in the top graph. One can pan through the entire signal by using the arrow buttons provided on the right of the graph. A click on "locate peaks" automatically detects the peaks by displaying a red vertical line in the signal. At this moment the second graph displays the time elapsed between two peaks (RR-interval in case of ECG signal) in chronological sequence. Two graphs in the bottom of the panel display values of beat-to-beat blood flow index and morphology index. Any inconsistent point in the lower three graphs can be clicked to get corresponding data in the top graph. Also, the small graph on the right displays the fast fourier transform (FFT) of the corresponding blood flow peak. Editing of the signal for "peak shift" or "peak insert/delete" can be done with the help of buttons provided on the right side of the panel. The processed and edited data is then saved in the same file. Click on quit brings back the application panel.

A click on display button displays HRV, PBFV and MIV spectra (top to bottom respectively) on the left side of the panel as shown in Figure 3. Left side of the graph gives time-domain variations, and that on the right gives frequency domain variations. VLF, LF and HF peaks are annotated in the HRV spectrum. The computed variability parameters are given in the corresponding windows on the right side of the panel. A click on "save to excel" transfers the data to a designated Excel sheet. One can quit this panel and go back to the application panel.

Mean and standard deviation (SD) values were calculated for 24 parameters in 5175 data files from 207 control subjects. Because of large values of SD, CLT was applied, which states that mean of a non-normally distributed variable computed from a large sample size is approximately normally distributed. Accordingly,

Table 2: Changes observed in variability parameters in different diseases

Sr No.	*Parameter*	*Deviation*	*Disease group*
1.	Total power in HRV	Drastic ↓	CAD
2.	HF center frequency in HRV	Mild ↑	Hypertension
3.	Total power in BFV	Moderate ↓	Diabetes
4.	HF amplitude in BFV	Moderate ↑	CAD and Diabetes
5.	Total power in MIV	Moderate ↓	Cancer
6.	HF amplitude in MIV	Moderate ↑	Hypertension
7.	LF amplitude in BFV	Moderate ↓	Hypertension and Arthritis
8.	HF center frequency in HRV	Mild ↑	Migraine
9.	HF amplitude in BFV	Mild ↓	Migraine

Table 3: Profile of subjects in various groups

Sr.No.	*Description*	*Control*	*TB*	*HT*	*IHD*	*AIDS*	*CoL*	*SC*	*LC*
1.	No. of subjects	207	46	23	31	31	21	20	35
2.	Age group	18–34	18–70	20–40	35–75	25–75	30–80	25–80	30–80
3.	Male/female	59/148	34/12	15/8	15/16	21/10	21/0	8/12	25/10

values of each parameter in a subject were averaged, and average values of 24 parameters in 207 subjects were taken for further statistical analysis. Mean and standard deviation values were thus computed using the above average values. The same procedure was applied to the patients in TB, HT, IHD, AIDS, CoL, SC and LC groups. For finding a significant difference between the control and disease groups, student's "t" test was applied to compare the yield of parametric statistical analysis. For comparison, Mann–Whitney U-test was applied on the raw (un-averaged) values of variability parameters and the Z-score value thus obtained was compared with "t" value to establish the relevance of parametric analysis in this data. Deviation of variability parameter value in the patient group with that of controls was measured in units of respective standard deviation value. For instance, a mean value of 33.838 in a patient group compared with 12.841 ± 6.489 (mean ± SD) of control group yield a deviation of 20.997, which is more than three times the standard deviation value in controls.

RESULTS

Table 4 gives mean and SD values of 24 variability parameters in control subjects and patients, after application of CLT. These parameters are:

Mean values of RR interval, PBF, and MI (RR_mean, BF_mean, and MI_mean);

Total power of HR, PBF and MI variations (HR_TP, BF_TP, and MI_TP);

Amplitude of VLF peak in HRV, PBFV and MIV (HR_aVLF, BF_aVLF and MI_aVLF);

Amplitude of LF peak in HRV, PBFV and MIV (HR_aLF, BF_aLF and MI_aLF);

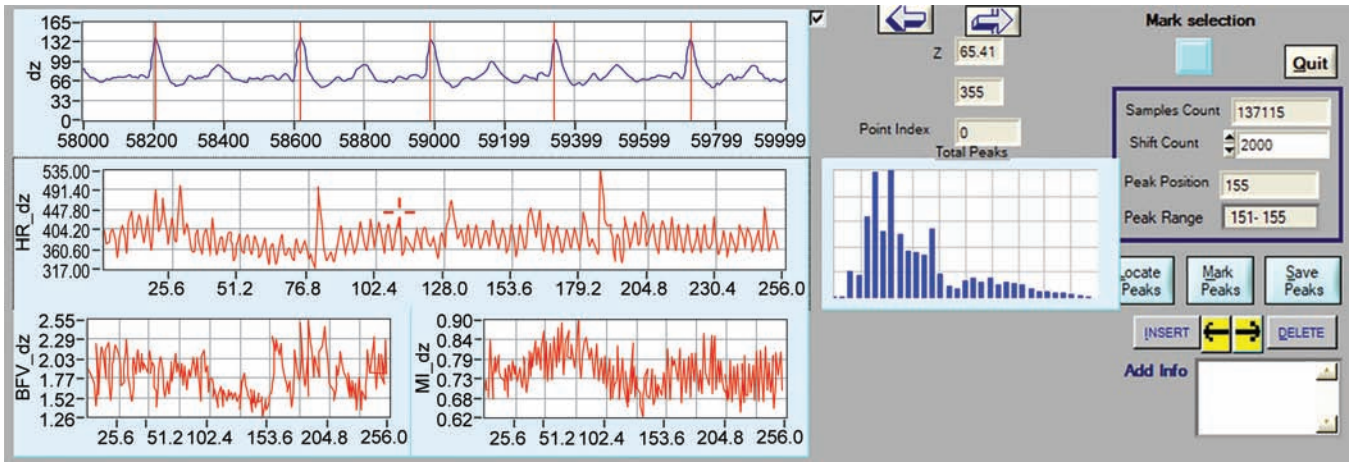


Fig. 2: GUI of the MARK panel. Top graph displays the raw signal. Three graphs below raw signal display beat to beat RR-interval, blood flow index and morphology index respectively. Editing of the peaks can be done with the help of buttons provided on the right side of the panel

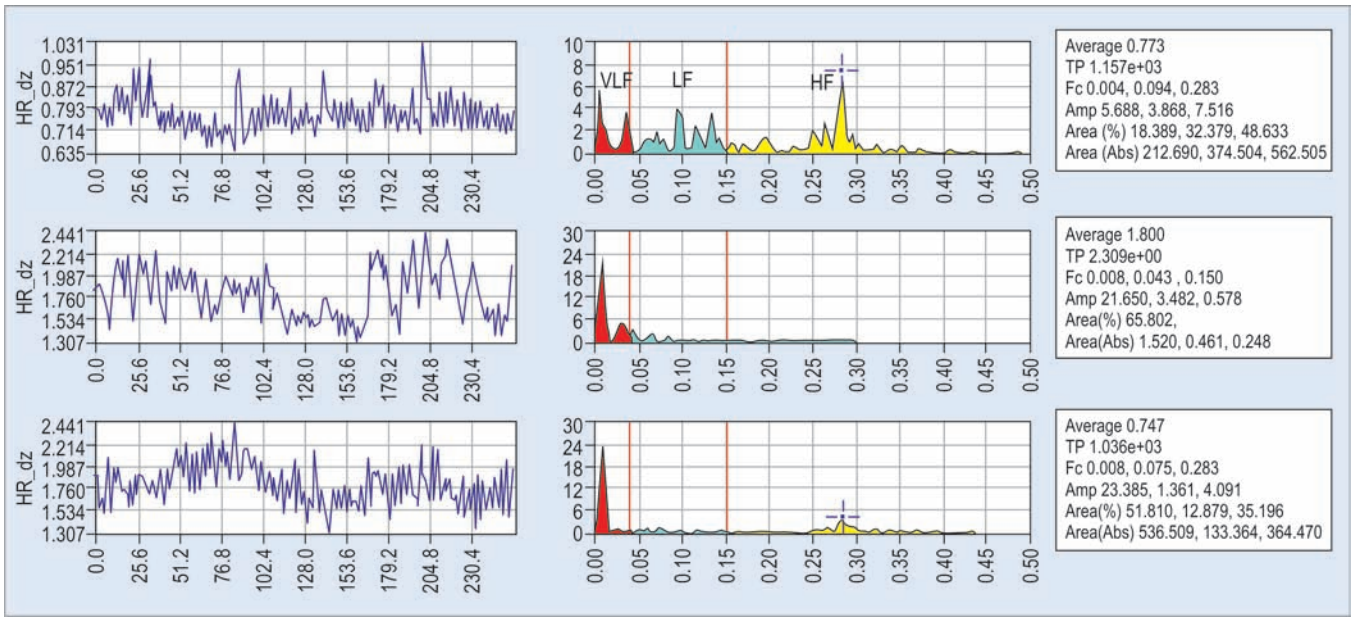


Fig. 3: GUI of the DISPLAY panel. Graphs on left display time domain variations in HR, PBF and MI respectively. Graphs on the right give corresponding frequency domain variations. Computed parameters are displayed in the three boxes on the extreme right. VLF, LF and HF are highlighted with red, blue and yellow color respectively

Table 4: Average and SD values of variability parameters after applying central limit theorem

	Control Subjects		% of	TB		HT		IHD		AIDS		CCOL		SC		LC	
Parameters	Mean	SD	Total SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
RR_mean	0.861	0.114	83.410	0.657	0.101	0.765	0.098	0.856	0.168	0.722	0.127	0.730	0.142	0.770	0.129	0.724	0.133
HR_TP	1435.1	953.4	71.114	229.5	374.9	286.7	234.0	456.0	521.1	302.5	439.9	171.0	180.5	249.8	275.5	268.1	533.9
HR_aVLF	11.028	3.440	46.777	15.489	7.865	15.425	6.565	16.762	6.079	14.367	6.523	15.377	8.537	16.343	7.250	12.736	6.850
HR_aLF	4.905	1.311	49.793	3.239	1.805	4.794	1.834	3.586	1.164	3.861	2.895	3.101	1.592	3.425	1.481	3.258	1.817
HR_aHF	4.214	1.924	57.928	2.944	2.886	2.473	2.127	3.014	2.434	2.664	2.206	3.256	2.460	3.350	2.933	3.988	2.986
HR_AVLF	28.226	9.223	56.185	46.267	17.238	44.239	12.979	48.275	15.077	45.013	16.113	40.814	21.055	45.591	16.113	38.126	17.731
HR_ALF	35.034	8.683	60.392	22.320	10.964	29.816	9.523	23.445	6.951	23.276	10.623	18.908	9.137	22.919	7.956	19.909	8.835
HRV_AHF	35.010	11.574	71.076	27.889	16.650	23.210	10.470	26.025	15.742	29.167	17.660	37.320	24.541	28.962	14.155	39.856	21.940
BF_mean	2.295	0.688	80.592	4.223	1.297	4.019	1.101	3.812	1.646	3.868	1.241	3.638	1.527	3.788	1.267	3.772	1.466
BF_TP	4.668	3.150	69.303	7.525	6.542	8.234	6.270	3.833	4.178	7.595	6.157	7.805	9.718	6.324	5.584	5.928	4.590
BF_aVLF	16.341	2.820	34.855	15.253	8.876	16.045	6.999	13.625	6.959	14.793	7.563	9.467	6.270	12.763	5.884	9.961	4.445
BF_aLF	7.054	2.101	48.842	3.693	2.216	3.538	1.583	4.045	1.705	3.430	1.385	3.141	0.531	3.535	1.182	3.801	1.745
BF_aHF	1.051	0.651	54.774	1.976	1.553	1.875	1.082	1.859	0.961	2.438	1.800	2.945	1.338	2.823	1.889	3.510	2.850
BF_AVLF	45.028	11.075	53.253	45.301	19.526	49.017	16.865	41.938	17.667	41.899	18.363	29.521	19.152	39.243	16.702	31.686	14.169
BF_ALF	40.017	10.452	56.668	23.109	9.158	21.324	5.565	25.740	7.646	22.087	5.833	23.227	4.427	23.388	6.438	23.059	5.295
BF_AHF	12.841	6.489	55.222	28.838	17.905	27.764	16.342	29.959	16.026	33.838	17.435	45.710	18.049	35.367	15.468	43.712	14.878
MI_mean	0.752	0.067	62.196	0.763	0.104	0.659	0.133	0.660	0.106	0.770	0.115	0.742	0.106	0.712	0.114	0.717	0.135
MI_TP	2007.5	1556.9	58.698	1143.7	983.7	3011.3	2532.6	989.1	668.3	1290.9	1163.4	1354.5	1348.4	1391.2	1196.7	1495.9	1223.3
MI_aVLF	17.609	4.116	40.642	15.586	8.077	18.550	9.016	15.651	9.244	14.911	8.160	10.398	6.599	15.067	7.794	11.720	6.212
MI_aLF	5.694	1.846	44.971	3.289	1.718	2.782	1.031	3.472	1.246	3.281	1.212	3.763	1.076	3.390	0.997	3.828	1.103
MI_aHF	1.534	0.824	51.616	2.036	1.421	1.934	1.529	2.188	1.349	2.350	1.164	3.209	2.268	2.693	2.393	3.228	2.334
MI_AVLF	46.562	11.926	51.840	45.460	18.583	50.749	18.687	44.343	21.304	41.338	17.319	29.807	15.904	40.467	18.054	34.869	17.096
MI_ALF	32.158	8.552	50.728	21.478	8.315	18.135	6.053	23.239	8.371	22.254	5.724	25.637	5.893	22.786	7.125	24.394	6.268
MI_AHF	18.655	8.474	57.761	29.776	13.736	26.550	17.417	29.438	16.724	34.274	14.933	43.036	15.756	34.222	15.533	39.347	14.852

Amplitude of HF peak in HRV, PBFV and MIV (HR_aHF, BF_aHF and MI_aHF);

Area of VLF peak in HRV, PBFV and MIV (HR_AVLF, BF_AVLF and MI_AVLF);

Area of LF peak in HRV, PBFV and MIV (HR_ALF, BF_ALF and MI_ALF); and

Area of HF peak in HRV, PBFV and MIV (HR_AHF, BF_AHF and MI_AHF).

The fourth column in the table expresses SD value (of the average value of parameters) as a percentage of raw SD value (of the raw data before averaging), which is ranging from 40.6 to 83.4% and signifies the effect of CLT. Table 5 shows values of student's "t," Mann–Whitney Z-score and deviation of patient group mean value from that of controls in units of respective standard deviation. Out of 168, 115 (68.45%), comparisons have shown a significant difference at a confidence level of 1% by student's "t." Z-score values obtained from non-parametric analysis using Mann–Whitney U-test have shown a significant difference at a confidence level of 1% in 125 (74.41%) comparisons out of 168. Figure 4 depicts a nearly linear relation between 't' and Z-score yielding a correlation coefficient of 0.8675.

As an additional measure, deviations of mean values of PV parameters in patients from that of controls in units of SD (control group) are obtained as given in Table 5. Twenty four out of 168 mean values are more than 2 SD away from the control mean and therefore suggest their discriminating potential. With these criteria, the majority of statistical significance is in BFV and MIV parameters; HRV has shown a change only in HR_AVLF in IHD. In general, BF_mean and BF_AHF are increased in all the diseases with exception of BF_mean in CoL. Therefore these are general parameters which increase in all the diseases and can be used as an index for post-therapeutic assessment. BF_aHF is explicitly increased in AIDS, CoL, SC and LC; suggesting a generalized increase in terminal diseases. Thus disease-specific parameters are HR_AVLF in IHD; BF_aVLF, MI_aHF, and MI_AHF in CoL and LC, which can be used for disease characterization.

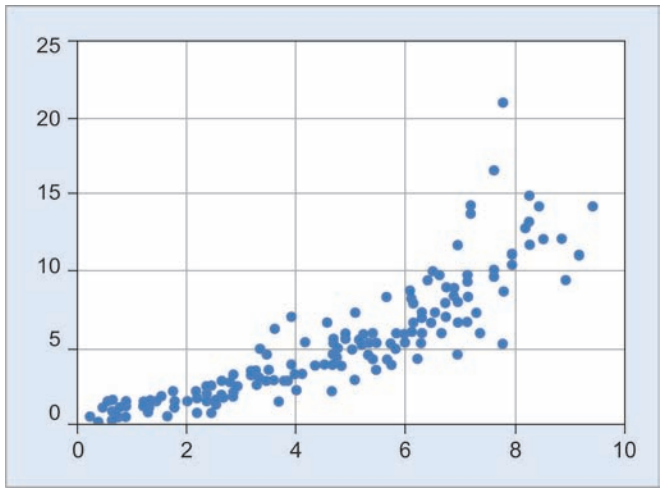


Fig. 4: Plot of student't' value (on Y axis) against Z score of Mann–Whitney U-Test (on X axis) for all the 168 comparisons in this study. The plot is satisfactorily linear up to Z score of 7.0 indicating that both the tests are leading to same conclusions. Correlation between the 2 parameters is computed to be 0.8675, suggesting compatibility between them

Table 5: Student's "t" values of comparison of patient's data with that of control subjects

	TB			*HT*			*IHD*			*AIDS*			*CoL*			*SC*			*LC*		
Parameter	*"t"*	*Z*	*nSD*	*"t"*	*Z*	*nSD*	*"t"*	*Z*	*nSD*	*"t"*	*Z*	*nSD*	*"t"*	*Z*	*nSD*	*"t"*	*Z*	*nSD*	*"t"*	*Z*	*nSD*
RR_mean	12.11	8.82	-1.00	4.42	3.58	0.00	0.17	0.70	0.00	5.75	5.85	-1.00	4.12	4.74	-1.00	3.05	4.11	-1.00	5.78	5.24	-1.00
HR_TP	13.97	9.42	-1.00	13.96	7.19	-1.00	8.54	6.93	-1.00	10.98	7.95	-1.00	16.40	7.61	-1.00	13.10	8.30	-1.00	10.42	7.95	-1.00
HR_aVLF	-3.77	4.55	1.00	-3.16	3.25	1.00	-5.13	5.47	1.00	-2.79	2.81	0.00	-2.32	3.27	1.00	-3.24	4.00	1.00	-1.45	1.30	0.00
HR_aLF	5.92	6.66	-1.00	0.28	0.73	0.00	5.78	4.97	-1.00	1.98	4.68	0.00	5.02	4.71	-1.00	4.31	5.42	-1.00	5.14	5.73	-1.00
HR_aHF	2.85	5.10	0.00	3.76	4.84	0.00	2.63	3.85	0.00	3.71	4.70	0.00	1.73	2.70	0.00	1.29	3.72	0.00	0.43	1.65	0.00
HR_AVLF	-6.88	3.92	1.00	-5.76	5.40	1.00	-7.20	6.56	2.00	-5.66	5.44	1.00	-2.71	3.32	1.00	-4.75	5.84	1.00	-3.23	3.48	1.00
HR_ALF	7.37	7.32	-1.00	2.51	2.50	0.00	8.36	6.19	-1.00	5.88	6.17	-1.00	7.74	6.14	-1.00	6.45	6.49	-1.00	9.39	7.15	-1.00
HR_AHF	2.76	3.81	0.00	5.07	4.22	-1.00	3.06	4.00	0.00	1.79	2.91	0.00	-0.43	0.64	0.00	1.85	2.68	0.00	-1.28	0.68	0.00
BF_mean	-9.78	7.66	2.00	-7.35	6.30	2.00	-5.07	6.00	2.00	-6.90	6.72	2.00	-3.99	4.67	1.00	-5.20	6.32	2.00	-5.85	5.81	2.00
BF_TP	-2.89	3.65	0.00	-2.69	3.33	1.00	1.07	2.58	0.00	-2.60	2.40	0.00	-1.47	1.26	0.00	-1.31	0.90	0.00	-1.56	1.52	0.00
BF_aVLF	0.82	2.24	0.00	0.20	0.65	0.00	2.15	2.92	0.00	1.13	1.86	0.00	4.97	5.30	-2.00	2.69	3.44	-1.00	8.22	7.16	-2.00
BF_aLF	9.39	8.98	-1.00	9.74	6.69	-1.00	8.87	6.70	-1.00	12.57	8.23	-1.00	20.98	7.80	-1.00	11.66	8.29	-1.00	9.88	7.67	-1.00
BF_aHF	-3.97	5.72	1.00	-3.58	3.54	1.00	-4.53	6.94	1.00	-4.25	5.69	2.00	-6.41	6.18	2.00	-4.17	6.26	2.00	-5.08	7.76	3.00
BF_AVLF	-0.09	0.43	0.00	-1.11	1.24	0.00	0.95	0.84	0.00	0.92	0.94	0.00	3.65	4.37	-1.00	1.52	2.56	0.00	5.30	5.26	-1.00
BF_ALF	11.03	9.22	-1.00	13.66	7.23	-1.00	9.19	6.37	-1.00	14.06	8.44	-1.00	13.89	7.22	-1.00	10.31	7.92	-1.00	14.71	8.28	-1.00
BF_AHF	-5.97	7.38	2.00	-4.34	4.65	2.00	-5.88	6.34	2.00	-6.64	6.99	3.00	-8.29	6.86	3.00	-6.46	7.15	3.00	12.08	8.56	3.00
MI_mean	-0.68	1.33	0.00	3.30	3.29	-1.00	4.73	4.80	-1.00	-0.84	2.44	0.00	0.46	0.30	0.00	1.55	1.84	0.00	1.53	0.63	0.00
MI_TP	4.77	3.41	0.00	-1.86	2.24	0.00	6.30	3.63	0.00	3.05	2.85	0.00	2.08	2.24	0.00	2.14	2.36	0.00	2.19	1.83	0.00
MI_aVLF	1.65	2.25	0.00	-0.50	0.23	0.00	1.16	1.80	0.00	1.81	2.64	0.00	4.91	5.00	-1.00	1.44	2.41	0.00	5.41	4.97	-1.00
MI_aLF	8.47	7.76	-1.00	11.63	6.98	-1.00	8.61	6.09	-1.00	9.55	7.14	-1.00	7.22	5.07	-1.00	8.95	6.88	-1.00	8.25	5.67	-1.00
MI_aHF	-2.31	3.00	0.00	-1.24	0.54	0.00	-2.63	2.68	0.00	-3.77	4.36	0.00	-3.36	5.52	2.00	-2.15	4.03	1.00	-4.25	6.24	2.00
MI_AVLF	0.39	0.93	0.00	-1.05	1.36	0.00	0.57	0.64	0.00	1.62	1.61	0.00	4.70	5.13	-1.00	1.48	2.10	0.00	3.89	3.97	0.00
MI_ALF	7.84	6.97	-1.00	10.05	6.52	-1.00	5.52	4.66	-1.00	8.34	6.05	-1.00	4.60	3.47	0.00	5.51	5.21	-1.00	6.39	4.58	0.00
MI_AHF	-5.27	5.98	1.00	-2.15	1.70	0.00	-3.52	3.19	1.00	-5.69	5.96	1.00	-6.99	6.31	2.00	-4.42	5.31	1.00	-8.02	6.77	2.00

DISCUSSION

A large number of studies have been conducted in the past to study the manifestation of various diseases on the variability spectrum .[2,6-17] Majority of the studies have followed the dictum that variability is sign of life. In general, there has been moderate to severe decrease in total power (HR_TP) and HF power in HRV (HR_AHF) in the presence of a majority of diseases. HRV has been observed to be an objective method for post-therapeutic assessment in post-cardiac transplant cases[11]and patients with cardiac failure[10]. It has also been proved to be a reliable method for early detection of diabetic peripheral neuropathy.[8,9] Its application in the majority of diseases has been limited because of difficulty in quantifying the changes in variability parameter owing to the large coefficient of variation (COV); nearly 100%. Nonparametric analysis, normally employed for such data, can yield a significant difference between the two samples but fails in the second step for quantifying changes in a particular subject. In the present work, CLT has been used by taking an average of multiple readings from control and patients to reduce the coefficient of variation. It has brought down the COV in the range of 40.6 to 83.41% and has also allowed the use of parametric statistics. This is in agreement with similar exercise carried out in past on pulse harmonic analysis.[18] Z-score values obtained from the nonparametric analysis (Mann–Whitney U-test) and student's "t" values have shown near linear relation up to Z value of 7.0. Though the graph has been divergent subsequently as seen in Figure 4, overall an excellent correlation (0.8675) with student's "t" has been seen. This justifies the use of CLT and parametric statistics, which has many advantages over the conventional nonparametric analysis.

In the past, BP variability has been studied more often than the BPF variability. However, studies have been much less in number in comparison to HRV because of the practical difficulties in collecting beat to beat BP data. Invasive BP monitoring is sparingly used for the obvious reasons. Noninvasive monitoring based on the principle of plethysmography is commonly used for variability study but its exorbitant cost prohibits the common use of the same. Ananthkrishnan et al.[2] has introduced peripheral blood flow variability and shown its variation in health and disease. Since recording PBF is simple and less expensive, it has been used in the present study.

Furthermore no additional effort is required for recording PBF, since its already being recorded for deriving HRV. Morphology index introduced by Jindal et al.[20] for early detection of coronary artery disease has also been included in this study. This is also derived from PBF and thus no additional signal need be recorded from the patient. Thus PBF recorded with the help of PPA instrument in one shot gives all the three variability signals, namely HRV, PBFV and MIV.

As described above one in HRV, four in BFV and two parameters in MIV are significantly different in patient's groups and show the importance of these newly introduced variabilities. BF_AHF is observed to increase in all the diseases in contrast to HR_AHF, which decreases in the presence of diseases. This shows that area under HF peak in the presence of disease has inverse manifestations in BFV in contrast to HRV. Diseases bring down HR_AHF but increase BF_AHF. This observation is similar to that observed in relation to baroreceptor sensitivity.[21,22] Baroreceptor sensitivity has been observed to correlate inversely with BP variability and positively with HRV. Because of this observation, BFV can be considered similar to BP variabilit and can be its affordable, practical non-invasive alternative.

In previous studies[14,15,17] IHD has shown a drastic decrease in HR_TP and a moderate increase in BF_aHF. However, no statistical significance was reported because of the large values of COV. In the present study significant decrease in these HRV parameters is observed, although not qualifying test of standard deviation. This deviation test has been introduced to identify specific parameters from statistically different 68% of the variability parameters. BF_AHF has shown significant increase, which is consistent with previous observation. Disagreement with previous observations is observed in patients with AIDS. Increase in BF_aHF has been recorded in contrast to the decrease observed earlier. Present results have statistical validity and therefore override previous observations.

In the past, HRV has been proved to be an effective method for investigating the sympathetic and parasympathetic function of the ANS in diabetes and post-myocardial infarction patients.[16,17] Spectral analysis of HRV has revealed the nature of diabetic autonomic neuropathy and also quantification of the benefits derived from corrective therapy in post-infarction patients. The present study has additionally shown the usefulness of BFV and MIV in a variety of disorders with statistical significance. With the criteria of two SD deviations, it may now be possible to quantify the severity of disease and benefits derived from corrective therapy.

CONCLUSION

Application of CLT has made a parametric statistical analysis of various parameters feasible. This may help in the quantification of the disease. Blood flow variability and morphology index variability have expressed higher sensitivity to diseases in comparison to well established HRV. Similarly, CoL and LC are characterized by an increase in MI_aHF and MI_AHF, which can be used in the management of these diseases. BF_mean and BF_AHF have shown moderate to the drastic increase in all the diseases and therefore can be used to differentiate disease from health.

ACKNOWLEDGMENTS

Authors are grateful to the Board of Research in Nuclear Sciences, Department of Atomic Energy, Government of India, for financially sponsoring research projects at Father Muller Homeopathic Medical College and Father Muller Medical College, Mangaluru; to the authorities of Electronics Division, Bhabha Atomic Research Centre (BARC), Father Muller Homeopathic Medical College (FMHMC), Father Muller Medical College, MGM Institute of Health Sciences and MGM's College of Engineering and Technology for providing all the help for smooth progress of the research work. They are also thankful to Dr Prathima, JRF working on BRNS Project at FMMC; and Dr Prasanna Kumar, and Dr Sajan, consultant homeopaths and former faculty members of Father Muller Homeopathic Medical College for their contribution in the above said BRNS research projects. Invaluable help from Shri Vineet Sinha, Scientific Officer (F), Electronics Division, BARC, for making the PPA instruments available for the projects; Smt. Gouri Sawant for processing high volumes of data; Smt. Jyoti V Jethe for her immense contribution during analysis is gratefully acknowledged.

REFERENCES

1. Hon EH, Lee ST. Electronic evaluations of the fetal heart rate. VIII. Patterns preceding fetal death, further observations. Am J Obset Gynecol 1963 Nov 15;87(6):814-826.
2. Ananthakrishnan TS, Pithawa CK. Introduction to physiological variability. In: Jindal GD, Deepak KK, Jain RK (eds.) A handbook on physiological variability. Mumbai: Electronics Division, Bhabha Atomic Research Centre; 2010. pp.1-16
3. Akselrod S, Gordon D, Ubel FA, Shannon DC, Berger AC, Cohen RJ. Power spectrum analysis of heart rate fluctuation: a quantitative probe of beat-to-beat cardiovascular control. Science, New Series 1981 July10;213(4504):220-222.
4. Pagani M, Lombardi F, Guzzetti S, Rimoldi O, Furlan R, Pizzinelli P, Sandrone G, Malfatto G, Dell'Orto S, Piccalgua E, et al. Power spectral analysis of heart rate and arterial pressure variabilities as marker of sympatho-vagal interaction in man and conscious dog. Cir Res 1986 Aug; 59(2):178-193.
5. Camm AJ, Malik M, Bigger JT Jr, Breithardt G, Cerutti S, Cohen RJ, Coumel P, Fallen EL, Kennedy HL, Kleiger RE, et al. Heart rate variability: standards of measurement, physiological interpretation and clinical use. Task force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology. Circulation 1996;93(5):1043-1065.
6. Acharya RU, Paul Joseph K, Kannathal N, Lim CM, Suri JS. Heart rate variability: a review. Med Biol Eng. Comput. 2006 Dec;44(12):1031-1051.
7. Jindal GD, Sawant MS, Pande JA, Rohini A, Jadhwar P, Naik BB and Deshpande AK. Heart rate variability: objective assessment of autonomic nervous system. MGM J Med Sci 2016;3(4):196-205.
8. Pagani M, Malfatto G, Pierini S, Casati R, Masu AM, Poli M, Guzzetti S, Lombardi F, Cerutti S, Malliani A. Spectral analysis of heart rate variability in the assessment of autonomic diabetic neuropathy. J Auton Nerv Syst.1988 Aug;23(2):143-153.
9. Bianchi A, Bontempi B, Cerutti S, Gianoglio P, Comi G, Natali Sora MG. Spectral analysis of heart rate variability signal and respiration in diabetic subjects. Med Biol Eng Comput 1990 May;28(3):205-211.
10. Copie X, Le Heuzey JY, Iliou MC, Khouri R, Lavergne T, Pousset F, Guize L. Correlation between time-domain measures of heart rate variability and scatterplots in postinfarction patients. Pacing Clin Electrophysiol 1996 Mar;19(3):342-347.
11. Sands KE, Appel ML, Lilly LS, Schoen FJ, Mudge GH, Cohen RJ. Power spectrum analysis of heart rate variability in human cardiac transplant recipients. Circulation. 1989 Feb;79(1):76-82.
12. Lutfi MF, Sukkar MY. Effect of blood pressure on heart rate variability. Khartom medical journal; 2011; 4(1); pp 548-553.
13. Natarajan N, Balakrishnan AK and Ukkirapandian K. A study of analysis of heart rate variability in hypertensive individual. International journal of biomedical and advance research; 2014;5(2);pp 109-111.
14. Jindal GD, Anathkrishnan TS, Mandlik SA, Sinha V, Jain RK, Kini AR, Naik MA, Kataria SK, Mahajan UA, Deshpande AK. Medical analyzer for the study of physiological variability and disease characterization. Navi Mumbai: Bhabha Atomic Research Centre; 2013. (External Report, BARC/2003/E/012).
15. Bhat S, Bhat KS, D'sa S, Roopa. Variability studies and disease characterization. In: Jindal GD, Deepak KK, Jain RK (eds) A handbook on physiological variability. Mumbai: Electronics Division, Bhabha Atomic Research Centre; 2010. pp. 101-105.
16. Ewing DJ, Winney R. Autonomic function in patients with chronic renal failure on intermittent haemodialysis. Nephron1975:15(6):424-429.
17. Varier PM, Muraleedharan K, Muraleedharan TS, Udayakumari TP, Sudheer AV. Evaluation of peripheral pulse analyzer from Ayurvedic viewpoint. In: Jindal GD, Deepak KK, Jain RK (eds) A handbook on physiological variability. Mumbai: Electronics Division, Bhabha Atomic Research Centre; 2010. pp. 128-136.
18. Jindal GD, Jain RK, Bhat Sushma N, Pande Jyoti A, Sawant Manasi S, Jindal SK and Deshpande Alaka K.Harmonic analysis of peripheral pulse for screening subjects at high risk of diabetes.Journal of Medical Engineering and Technology 2017;41(6):437-443.
19. Deepak KK, Jindal GD. Draft protocol for recording short term physiological variability.In: Jindal GD, Deepak KK, Jain RK (eds) A handbook on physiological variability. Mumbai: Electronics Division, Bhabha Atomic Research Centre; 2010. pp. 144-149.
20. Jindal GD, Jain RK, Sinha Vineet, Mandlik SA, Sarade Bhagyashree, Tanawade Pooja, Pithawa CK, Kelkar PM and Deshpande AK. Early detection of coronary heart disease using peripheral pulse analyzer. BARC Newsletter 2012;326:15-21.
21. Mancia G, Parati G, Pomidossi G, Casadei R, Rienzo MD, Zanchetti A. Arterial baroreflexes and blood pressure and heart rate variability in humans. Hypertension 1985;8(2): 147-153.
22. Hesse C, Charkoudian N, liu Z, Joyner MJ, Eisenach JH: Baroreflex sensitivity inversely correlates with ambulatory blood pressure in healthy normotensive humans. J Hypertension 2007;50:41-46.