

## Evaluation of Cardiac Responses to Stress in Healthy Individuals - A Non Invasive Evaluation by Heart Rate Variability and Stroop Test



## Medical Science

**KEYWORDS :** Heart rate variability, Stroop test, Autonomic nervous system.

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### ABSTRACT

*Objective and Study Design:* Stress is considered as physiological response to the mental, emotional, or physical challenges. Most of the times mental stress provokes body's "fight or flight" response called as acute stress response. But prolonged or chronic stress can affect numerous physiological functions, such as growth, immune system, metabolism, reproduction and circulation. The Autonomic regulation of the cardiovascular system is most commonly affected by stress and is assessed by means of heart rate variability (HRV)

*Material and Methods:* The aim of this study is to establish mental stress and cardiovascular autonomic Nervous System response to mental stress assessed by combining different physiological parameters using HRV and Stroop test as tool. We compared the mean RR interval, Blood pressure and indices of HRV during the Stroop Color Word Test (SCWT). A total of 50 normal healthy subjects were participated in this study.

*Results:* Statistically significant change in heart rate, RR interval and BP was observed during the stress as compared to resting condition. All the components of HRV (SDNN, RMSSD, NN50, PNN50, LF, HF, LF/HF) were sensitive to stress in all the healthy individuals. Males were more prone to stress when compared to females.

*Conclusion:* The results suggest that there was increased sympathetic activity and reduced parasympathetic activity in HRV performed during the Stroop test and showed statistically significant difference among the genders. Both physical and mental stress influence risk factors that may increase risk for cardiovascular diseases and autonomic imbalance

### INTRODUCTION:

Stress is a huge problem in today's society and is an inevitable part of everyday life. Although stress has a psychological origin, it affects several physiological processes in the human body. Physiological stress affects homeostasis, leading to increase in the production of cortisol by the hypothalamic-pituitary adrenal axis (HPAA) and changes in the cardiovascular system brought about by the Autonomic Nervous System (ANS). On the other hand, psychological stress causes bodily changes that originate in the higher centers of the brain and act in the periphery via the ANS. The brain innervates the heart by means of stimuli via the Autonomic Nervous System (ANS), which is divided into sympathetic and parasympathetic branches. The sympathetic activity leads to an increase in HR (e.g. during sports exercise), while parasympathetic activity induces a lower HR (e.g. during sleep). The two circuits are constantly interacting and this interaction is reflected in HRV. HRV, therefore, provides a measure to express the activity of the ANS, and may consequently provide a measure for stress. Though autonomic response to stress has been assessed using different techniques like Plasma and urinary catecholamine levels, cardiac norepinephrine spillover and micro-neurographic techniques, the concept of Heart Rate Variability (HRV) has been proposed as a robust, non – invasive and sensitive tool to study the influence of both sympathetic and parasympathetic systems on the heart. Currently, the time and spectral analysis of HRV is seen as a research and clinical tool for the study of cardiovascular autonomic regulation. Short term HRV has been used as a quantitative probe to assess the modulation of heart rate by the Sympathetic Nervous System (SNS) and the Parasympathetic Nervous System.

Numerous stressors have been developed to study the autonomic modulation of cardiovascular function in the laboratory like stress ergometry, exercise, cold pressor test or Valsalva manoeuvre. The hemodynamic response to mental stress differs from the corresponding response to physical

exercise. So we preferred stroop test as a stressor. Stroop test is commonly used as a mental stressor in cardiovascular research. It is classically called as Stroop color word test which was originally introduced by John Ridley Stroop. The Stroop Color and Word Test is based on the observation that individuals can read words much faster than they can identify and name colors. Its quick and easy administration, validity, and reliability make it a highly useful instrument. Essentially it comprises of three kinds of stimuli: colored rectangles serving as the control, names of colors written in the congruent color (eg. 'GREEN' in green) and the names of colors presented in an incongruent color ('GREEN' in red). The test requires the subjects to name the color of the rectangles, the words of the congruent stimuli and the color of the incongruent stimuli, without reading the actual word itself. When presented with the incongruent stimuli, the subject gets into a conflict filled stressful situation because his reply is influenced by his learning. This is referred to as the Stroop interference. Among all mental stressors, only the Stroop test has been found to activate all components of the SNS.

Our study was inspired by the fact that very few studies were done in Indian subcontinent to demonstrate how different individuals react to physical as well as mental stress and document its effect on HRV. Several Studies have shown that the computerized Stroop test causes an increase in heart rate, decreases HRV and increases catecholamine secretion thus making it a suitable and well standardized mental stressor. In the current study HRV recordings were taken during rest and stroop color word test and studied how cardiovascular reactivity to stress is associated with changes in HRV parameters.

### Materials and Methods:

Fifty healthy young volunteers were included in the study who were assessed for HRV and Stroop test after acquiring oral and written consent. The study also got the approval from ethical committee, AIIMS, Bhopal. None of the subjects had been previously diagnosed with cardiovascular or

psychiatric disorders, nor were they taking any medication that would alter their heart rate. Subjects who had taken a Stroop Test earlier and those who were color blind or dyslexic were excluded from the study. Volunteers were instructed not to eat a heavy meal, ingest caffeine or alcohol or exercise 4 hours before arriving at the laboratory. They were also asked to void urine before the test.

Heart rate, Respiratory rate and baseline blood pressure was recorded for all subjects. The computerized version of the Stroop color word test from ADInstruments Power lab was used as a mental stressor and the subjects were trained to respond using a keyboard. They were instructed to answer as quickly and accurately as possible. Initially ECG electrodes were connected to the volunteers and ECG was recorded for 5 minutes after 15 minutes of rest at comfortable room temperature. After which each of them were subjected to stress by means of Stroop test and ECG was recorded for 5 minutes. The acquired ECG signal during rest and Stroop test is digitized, stored and analyzed into time and frequency domain methods of HRV by ADInstruments power lab.

**Stroop test:**

The easy practice test: just name the color (not the word says)



The real hard test: Name the Color (not what the word says). (Note that it is the same list of words but written in different color.) (PAY ATTENTION: The Color of the word is different from what the word says.)



It clearly stated that, it takes much longer time to complete the second test than the first test. The first test is easy because the color and meaning of the word are congruent. There is no conflict. The second test is hard because the color and meaning of the word are incongruent. This cre-

ates a conflict that the brain has to resolve. The reason why it takes longer is because the brain has to suppress the wrong answer that interferes with the right answer, before the right answer comes through.

**HRV:** The frequency domain and time-domain indices which were reported from the HRV recordings include:

**Time domain**

- Mean heart rate
- Mean RR
- SDNN - Standard deviation of all R-R intervals (ms)
- RMSSD - Square root of the mean of the successive differences between R-R intervals (ms)
- NN50 - number of interval differences of successive NN intervals greater than 50 ms
- PNN50 - Percentage of successive differences between R-R intervals greater than 50 ms (%)

**Frequency domain**

- TF - Total power ( $\mu\text{s}^2$ )
- VLF - Very Low frequency spectrum ( $\mu\text{s}^2$ ) (Between 0 and 0.04 Hz)
- LF - Low frequency spectrum ( $\mu\text{s}^2$ ) (Between 0.04 and 0.15 Hz)
- HF - High frequency spectrum ( $\mu\text{s}^2$ ) (Between 0.15 and 0.45 Hz)
- LF/HF - LF/HF ratio
- Normalized LF and HF

**Results:**

Results were analyzed by SPSS version 17.0 and Graph pad prism. ANOVA and Unpaired Student,  $t_{\text{test}}$  test was used to compare the data. All values were expressed as Mean $\pm$ SD. The P values < 0.05 were statistically considered significant.

TABLE I: Shows Anthropometrical measurements in all healthy volunteers and Comparison of respiratory rate, systolic blood pressure (SBP) and diastolic blood pressure (DBP) and mean arterial pressure (MABP) of the subjects during rest and during stroop test. Data presented are Mean $\pm$ SD. The data analyzed using students unpaired t-test to compare the mean values between two groups. There was no statistically significant difference in DBP and MAP in individuals during rest and Stroop test. However Immediately after the test the increase in SBP and respiratory rate values were statistically significant.

	Hrv analysis in participants during rest	Hrv analysis in participants during Stroop test	P value
Age			
Height (cms)	28.52 $\pm$ 0.71		
Weight (kg)	173.55 $\pm$ 4.96		
BMI	65.96 $\pm$ 7.89		
	22.63 $\pm$ 1.96		
Respiratory rate	14 $\pm$ 1.00	16 $\pm$ 0.96	0.0001***
SBP (mmhg)	115.86 $\pm$ 7.63	120.32 $\pm$ 5.63	0.0012**
DBP (mmhg)	80.98 $\pm$ 6.12	81.69 $\pm$ 7.23	0.5973
MABP (mmhg)	91.49 $\pm$ 5.36	93.62 $\pm$ 5.94	0.0627

TABLE I I: Shows comparison of Time domain measures of HRV in individuals during rest and stroop. We found that there is no statistically significant difference in mean R-R interval and NN50, PNN50% in the individuals during rest and stroop test. However mean HR, SDNN, RMSSD values showed statistically significant difference during rest and Stroop test.

	Time domain parameters in participants during rest	Time domain parameters in participants during Stroop test	P value
Mean HR	80.20 ± 13.26	86.14 ± 10.36	0.0142*
Mean RR	770.90 ± 113.59	745.90 ± 114.36	0.2754
SDNN	69.27 ± 22.06	53.36 ± 26.36	0.0015**
RMSSD	51.44 ± 23.10	39.51 ± 25.69	0.0164*
NN50	49.63 ± 33.18	91.76 ± 70.46	0.6502
PNN50%	26.23 ± 17.45	25.61 ± 16.80	0.8567

TABLE I I I: Shows comparison of Frequency domain measures of HRV in individuals during rest and stroop. In our study, Total Power, LF nu, HF nu, showed no significant difference among participants during rest and stroop test. However, during the stroop test, there was a statistically significant increase in LFms2, LF-HF ratio and decrease in HFms2 when compared to resting participants.

	Frequency domain parameters in participants during rest	Frequency domain parameters in participants during Stroop test	P value
TP	3641.74 ± 2517.86	2936.36 ± 1998.89	0.0511
LF ms <sup>2</sup>	1257.26 ± 955.63	1360.48 ± 752.11	0.0261*
HF ms <sup>2</sup>	952.36 ± 745.39	849.39 ± 649.36	0.0463*
LF nu	54.29 ± 14.39	56.01 ± 10.26	0.4930
HF nu	43.63 ± 15.59	40.59 ± 10.28	0.2525
LF/HF	1.19 ± 0.51	2.21 ± 0.89	0.0001***

### Discussion:

HRV analysis has gained much importance in recent years as a technique employed to explore the activity of ANS, and as an important early marker for identifying both physiological and pathological conditions. An attempt was made to examine the influence of stress on Heart Rate Variability. The objective of the study was to evaluate the cardiovascular responses to stress induced by the computerized version of the Stroop word-color test. The results of the physiological measurements of HRV during stroop test proved increased sympathetic activity in individuals during the stroop test. Also, significantly higher systolic blood pressure was found in stress group i.e. stroop test. This may be due to the changes in the heart rate which raises the SBP and the fact that the diastolic pressure is mainly due to peripheral resistance, which is unlikely to be altered. Several studies have shown that there appears to be a correlation between stress, the HPA axis and the ANS control of the cardiac activity. When the person is exposed to stress, hormones play a role in decreasing vagal and increasing sympathetic activity by enhancing the cholinergic muscarinic activity at central and peripheral levels. It has been postulated that both physiological and psychological stress contributes to the change in blood pressure, HR, HRV and reported significantly higher SBP and increased sympathetic activity in stress group when compared to rest group. This strongly suggests that there is activation of the sympathetic adrenal medullary axis by stress. Other studies have also shown higher heart and respiration rates during mental and physical stress than at rest.

Mean values ± SD for heart rate increased highly and significantly during physical stress. Thus, as expected there was significant decrease in RR interval both during stress. SDNN measures the overall HRV and estimates changes in heart rate due to cycles longer than 5 min. Low SDNN indicates low HRV which is not good for healthy life. In our study, significantly lower SDNN values in stress groups in-

dicating stress affects the cardiac autonomic function and increase the risk. RMSSD estimates high frequency variation in heart rate in short term recordings. Low RMSSD along with Low SDNN indicates cardiac risk. The same reflected in our study as well. NN50 and PNN50% estimates short term HRV and reflects vagally mediated response of heart rate. The values are not statistically significant between rest and stress group. LF ms<sup>2</sup> indicates mainly the sympathetic nervous system and also the parasympathetic nervous system to some extent whereas HF ms<sup>2</sup> indicates the activity of parasympathetic nervous system. In our study, significantly high LF and Low HF values were seen in stress group when compared to rest group clearly proved that increased sympathetic and decreased parasympathetic activity is seen during stress. LF/ HF indicate sympatho – vagal balance which showed statistically significant higher values in stress group than the rest group. LF/HF ratio less than 2 indicates healthy stress levels.

In our study, a significant difference was observed between the rest group and stress group with respect to their overall HRV status. It was found that participants underwent for stroop test had significantly reduced heart rate variability throughout the test in the form of decreased vagal and increased sympathetic activity. This was reflected by lower total power, HF ms<sup>2</sup>, HF nu and increased mean heart rate, LF ms<sup>2</sup>, LF nu, LF/HF ratio in individuals during rest when compared to stress. Moriguchi et al and Lucini D et al also documented the same.

### Conclusion:

In conclusion, our results are in support of higher sympathetic activity and lower parasympathetic activity in response to stress test (stroop test). Thus, we conclude both physical and mental or any kind of stress influence risk factors that may increase risk for cardiovascular diseases.

### Scope for further study:

Scope and reliability of the study can be enhanced by:

- HRV was recorded for a very short duration. A 24 hours Holter recording for HRV would be a better choice to understand the cardiovascular autonomic activity
- Increasing number of subjects.
- Including subjects from different professions.
- Testing efficacy of other physical and mental stress inducers, to identify the optimal inducer to be used for the study.
- Assessing biochemical parameters like cortisol and catecholamines during the study.

### ACKNOWLEDGEMENTS:

Each author should have participated sufficiently in the work to take the public responsibility for appropriate portions of the content. All authors read and approved the final manuscript. We are thankful to subjects and all the technical staff for their contribution in the completion of the project.

### CONFLICTS OF INTEREST: Nil

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