International Journal of Life Sciences, Biotechnology and Pharma Research Vol. 13, No. 8, August 2024 Online ISSN: 2250-3137
Print ISSN: 2977-0122

DOI: 10.69605/ijlbpr_13.8.2024.71

ORIGINAL RESEARCH

The correlation of serum inflammatory and oxidative stress markers in pre and stage I hypertension to predict pre-hypertension

[1]Sohana Fatma, [2]Dr. Anil Kumar Munta, [3]Dr. D Lakshmi Lalitha, [4]Dr. D SSK Raju

[1]MBBS Intern, Madhubani Medical College, Madhubani, Bihar, India
[2]Professor, Department of Biochemistry, Madhubani Medical College, Madhubani, Bihar, India
[3]Principal & Professor of Biochemistry, Great Eastern Medical School, Ragolu, Srikakulam,Andhra Pradesh,India
[4]Associate Professor,Department of Biochemistry, Maharajah's Institute of Medical Sciences, Nellimarla, Vizianagaram, Andhra Pradesh, India

Corresponding Author
Dr. D SSK Raju
Associate Professor,Department of Biochemistry, Maharajah's Institute of Medical Sciences, Nellimarla, Vizianagaram, Andhra Pradesh, India

Received: 25June, 2024 Accepted: 23July, 2024

ABSTRACT
Blood pressure is the force exerted by circulating blood against the arterial walls. The elevation of the pressure is known as hypertension and is one of the major global health concerns. Early diagnosis and timely interventions are essential and in predicting prehypertension to include as regular panel marker, this current work correlated the biomarkers in normal, pre and stage-I hypertensives to set the risk ratio and cut-off values independently. The informed consent was obtained from all the age and gender matched participants from the respective groups i.e., control group, pre-hypertensive and hypertensive groups. The age group of 30 to 60 years were chosen for the study. All the individuals FBS, Fibrinogen, Malondiadehyde and High sensitive C-reactive protein (hSCRP). Among the parameters, fibrinogen, MDH and hsCRP, the serum hsCRP have showed slight elevation the pre hypertension group compared to control and can be treated as predictive marker provided by analysing the more sample size. The serum fibrinogen, MDH and hsCRP have shown an increase in the stage I hypertension group compared to control and pre HTN which are statistically significant. The biomarkers showed slight association in predicting the hypertension.

Key words: Hypertension, prehypertension, fibrinogen, malondiadehyde and high sensitive c-reactive protein

INTRODUCTION

Blood pressure is the force exerted by circulating blood against the arterial walls. The elevation of the pressure is known as hypertension and is one of the major global health concerns. In 2015, nearly 1.13 billion people globally having hypertension and less than one in five is under control and expected to reduce to 25% by 2025 [1]. However, several studies reporting that the prevalence of hypertension is increasing gradually in an uncontrolled manner and it is the third most important risk factor for disease complications especially in south Asia compared to rest of the world. It is treated as "silent killer" as it is a significant risk factor for stroke, peripheral vascular diseases, renal disorders and coronary diseases. The decrease blood flow and oxygen supply to the heart is due to hardening of arteries in hypertension. The prehypertension, is an initial warning alarm and majority of the population are unaware of their disease condition. If the disease is not diagnosed in time and preventive measures are not taken then prehypertension soon gets converted to hypertension. There are several stages in hypertension and are often silent and asymptomatic [2].Approximately 90% of hypertensive patients suffer from essential or primary hypertension (EH), this is the condition when the cause of blood pressure (BP) elevation is unknown. Therefore, the regular monitoring of blood pressure is crucial to curtail the disease-oriented complications. The prevalence of hypertension increases with progressing age affecting both male and female population. Framingham Heart Study indicates that about 90% of people over the age of 55years will gradually develop hypertension [3].

DOI: 10.69605/ijlbpr_13.8.2024.71

On prevention, detection, evaluation and treatment of high Blood Pressure (JNC7), the Seventh Report of the Joint National Committee published in 2003 uses the term prehypertension for blood pressure in the range 120-139 mmHg systolic or 80-89 mmHg diastolic. Hypertension is classified as hypertension stage I, hypertension stage II and isolatedsystolic hypertension. Sometimes it is difficult to identify the pre hypertension or hypertension until it is elevated on several episodes and diagnosed only by regular monitoring of the BP readings [4]. The abnormal or fluctuating BP readings should be monitored carefully and the single BP reading is not able to predict the condition [2, 5]. In hypertension, the vascular function is affected and due to remodelling several biomarkers are released from vascular endothelium (increased media width, reduced lumen size and vascular resistance etc.) [6]. The utilization of reliable biomarkers to diagnose, predict and tracking the therapeutic progression of hypertension is proving to be tremendously significant. However there are several diversified opinions on the elevation of serum markers in hypertensionas they are release in the serum is influenced by certain factor apart from vascular dysfunction and it still remains undetermined to choose an early diagnostic marker for diagnosing prehypertension. The inflammatory markers and oxidative stress markers will rise in hypertension and associated with its development [7].

There is paucity and variety of literature regarding biomarkers in forecasting its strongest association in hypertension. Therefore, the biomarkers to be focused by considering the factors like specificity, easy versatility, availability and cost affectivity. It is still challenging and a mix up to the early and accurate diagnosis of pre and hypertension and any chronic condition that leads to devastating consequences. Early diagnosis and timely interventions are essential and in predicting prehypertension to include as regular panel marker, this current work correlated the biomarkers in normal, pre and stage-I hypertensives to set the risk ratio and cut-off values independently.

The latest studies have shown the generality of hypertension is 25% in urban and 10% in rural population in India and the disturbing trend is that it mostly affecting the young adults (20-44 years) [8]. Poor awareness, diagnosis, treatment and control are worsening the situation more [9].There is consistent relation between increased BP and vascular abnormalities and independent from other risk factors. The higher the BP, the greater is the risk of cardiac ailments, stroke, renal impairments etc.[10]. The blood pressure is measured as systolic and diastolic pressures, the maximum and minimum pressures [11]. However, there are certain cons in measuring the BP by manometers like position of an individual, patient movement, cardiac arrhythmias, rapid pressure changes, inappropriate cuff size, technical errors, lacks of expertise, severe shock, abnormal heart rate and hyperlipidemias [12].

The serum markers are having the prominent role in diagnosing and predicting the HTN. These biomarkers are increased in vascular dysfunction, inflammation and oxidative stress. The rapid degradation of NO by the oxygen-derived free radical superoxide anion modifies endothelial function. If oxidative damage is indeed a cause of hypertension, thendepletion of oxidative damage to tissues may result in reduced blood pressure [13, 14]. Fibrinogen is a plasma glycoprotein, plays an important role in inflammatory response and is a positive acute phase protein [15]. The degree of oxidative stress can be measured by the circulatory of levels of Malondialdehyde [16,17]. CRP promotes phagocytosis by activating complementary system and clears the apoptotic, necrotic cells and pathogens [18].

AIM AND OBJECTIVES

The aim of the present study is to assess the correlation of inflammatory and oxidative stress markers with normal, pre and stage I hypertensive cases to predict the hypertension.

- To evaluate fibrinogen, high sensitive C-reactive protein (hsCRP) and malondialdehyde in individuals with pre and stage I hypertensive stages. The current study may provide the necessary analytical report regarding these markers in correlation with the respective hypertensive stages.
- To compare and to set risk ratio among the fibrinogen, hsCRP and malondialdehyde in normal and stage I hypertensive individuals as an indicator in assessing the disease.
- To determine the predictive element by establishing cut off values for these markers independently.

MATERIALS AND METHODS SELECTION OF PARTICIPANTS

This descriptive study was carried out after approval of the Institutional Ethics Committee. The informed consent was obtained from all the age and gender matched participants from the respective groups i.e., control group, pre-hypertensive and hypertensive groups. The age group of 30 to 60 years were chosen for the study.

INCLUSION CRITERIA AND EXCLUSION CRITERIA

The participants of the corresponding groups (n=40 cases) who have fulfilled the diagnostic criteria prehypertension and hypertension as per the JNC7 guidelines were included. Individuals with smoking, alcoholism, obesity, pregnancy, cardiac ailments, thyroid disorders, renal impairment was excluded from the study as these factors influence the blood pressure.

DOI: 10.69605/ijlbpr_13.8.2024.71

METHODS: ESTIMATION OF BIOCHEMICAL PARAMETERS

FASTING BLOOD SUGAR (FBS): It was estimated by glucose oxidase and peroxidise method. Serum Fibrinogen measured by two site enzyme linked immunoassay (ELISA). The quantity of the fibrinogen in the test samples from standard curve.

ESTIMATION OF MDA: Assayed with Thiobarbituric acid by Keisatoh method. The lipid peroxide content is estimated as MDAequivalent of TBA assay values which measures total MDA (free and unbound). High sensitive C-reactive protein (hSCRP): It was measured by ELISA method.

SAMPLE COLLECTION

A detailed clinical examination was recorded with reference to age, sex, family and clinical history from all the participants. Considering the standardized protocol in the different stages of hypertension, the participant's blood pressure was measured with the aid of clinician by using mercury column sphygmomanometer. The standardized protocol involved in measurement of systolic and diastolic blood pressures, the average of two systolic and diastolic blood pressure measurements was obtained at the examination [19]. The bloodsample was collected separately, 2ml will be taken in EDTA vacutainers for fibrinogen and 3ml were collected to plain vacutainers for the analysis of other parameters. These blood samples in the respective tubes were separated and preserved at -40^{o}C for batch wise analysis of parameters in the laboratory.

STATISTICAL ANALYSIS

All the statistical analysis was carried out by using SPSS (Statistical Package for Social Sciences) trial version 16 in MS excel 2007. The quantitative variables were expressed as Mean and Standard deviation. $p<0.05$ is considered as statistically significant.

Kruskal Wallis test was used for mean comparison between the groups and receiver operator characteristic (ROC) curve was used to identify the optimal cut off values.

RESULTS

The descriptive and mean comparison between the groups is shown in Table 1. It is observed that there is a statistical significant difference between the levels of fibrinogen, hsCRP and Malondialdehyde level in Pre hypertension and Stage 1 Hypertension group when compared to control. The receiver operator characteristic (ROC) curve was plotted for Pre hypertension and Stage 1 HTN shown in Figure 1 and Figure 2. The value of cut off for markers are shown in table 2 and table 3 of pre and stage I HTN.For pre Hypertension the cut off levels for Fibrinogen, Malondialdehyde and HsCRP are respectively 366,3.5 and 1.1. It has been noticed from the findings that in stage I hypertension the cut off levels for fibrinogen 425 with sensitivity of 97.5% and Specificity of 97.5%. Malondialdehyde is 5.3 with sensitivity of 97.3% and specificity of 96.3% and hsCRP level 2.75 with sensitivity of 95% and Specificity of 95%.

Table 1: Comparison of Mean betweenGroups

Variable	Control Group	Pre Hypertension	Stage-1 Hypertension	KW-H/df/pvalue
Age Mean± SD	44.53 ± 8.8	43.98 ± 8.8	46.70 ± 9.0	2.0/2/0.3
FBS Mean± SD	79.6 ± 5.2	80.9 ± 5.2	84.1 ± 7.0	8.4/2/**0.015**
Fibrinogen Mean± SD	279.4 ± 43.5	395.2 ± 18.1	480.8 ± 28.5	**105.6/2/<0.001**
Malondidehyde Mean ± SD	2.1 ± 0.5	4.9 ± 0.2	5.7 ± 0.2	**103.7/2/<0.001**
hsCRP Mean ±SD	0.6 ± 0.18	2.0 ± 0.5	4.2 ± 0.48	**105.9/2/<0.001**

DOI: 10.69605/ijlbpr_13.8.2024.71

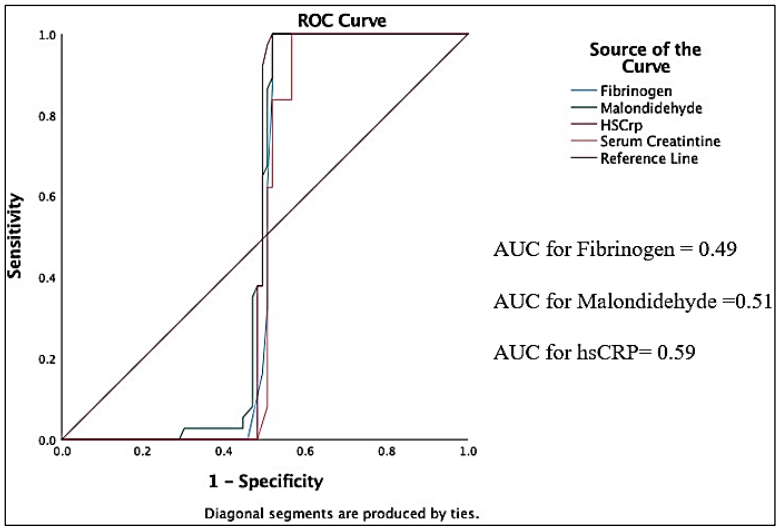

Figure 1:ROC curve for Diagnosis of Pre Hypertension

- **ROC:** Receiver Operator Characteristic curve.
- **AUC:** Area under the Curve.

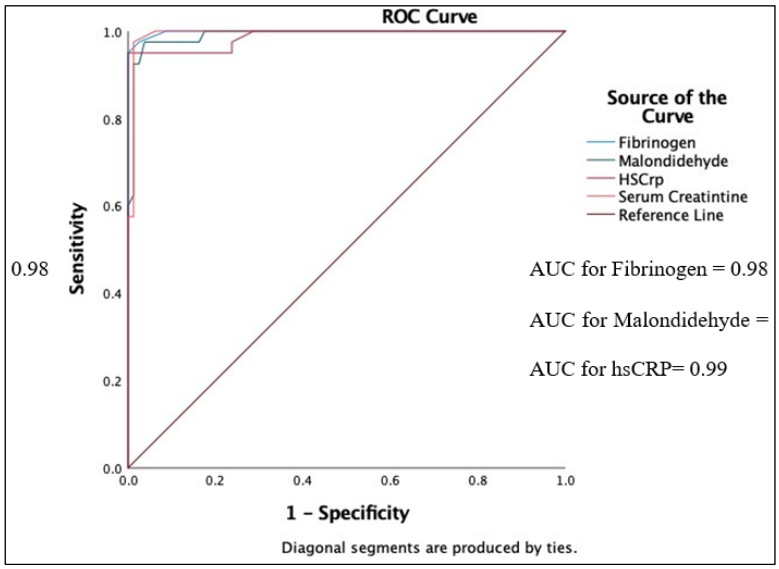

Figure 2: ROC Curve for Stage 1 Hypertension

- **ROC:** Receiver Operator Characteristic curve.
- **AUC:** Area under the Curve.

Table 2: Biomarkers cut off for prehypertension and Sensitivity and Specific

Biomarker	Cut off level
Fibrinogen	366 mg/dl
Malondidehyde	3.5 nmol/ml
hsCRP	1.9 mg/l

- HsCRP-high sensitive C reactive protein.

Table 3: Biomarkers cut off for stage I hypertension and Sensitivity and Specificity

Bio Maker	Cut off Level	Sensitivity	Specificity
Fibrinogen	425 mg/dl	97.5%	97.5%
Malondidehyde	5.3 nmol/ml	97.3%	96.3%
hsCRP	2.75 mg/l	95%	95%

DOI: 10.69605/ijlbpr_13.8.2024.71

According to the findings obtained from the Table I the markers hsCRP, fibrinogen and malondialdehyde were increased in pre and stage I HTN in compared to control which are statistically significant. Studies claim that the inflammatory and oxidative stress markers will be elevated in both the pre and stage-I HTN which is due to vascular endothelium dysfunction. The CRP preferentially elevated in stressed vessels. Studies reported that hsCRP is surprisingly specific for the prediction of vascular events [20, 21]. Malondialdehyde is significantly increased in both the study groups and the rise of MDH in pre and Stage I HTN can be accounted by increased oxidative stress which alters its level. Studies have shown that MDH is a potential marker of recognising pre HTN, however it lacks specificity. In one study the MDH in combination with other Oxidative stress markersprovides a better diagnostic and prognostic marker but not alone serves as diagnostic marker and not specific for the HTN. There is increasing evidence that free radicals are involved in the pathogenesis of hypertension by altering endothelial function [16]. The inflammation marker, plasma fibrinogen levels were raised significantly in both the experimental groups in the current study. Several studies showed that the positive association between fibrinogenemia and HTN. The structural alterations in the artery wall including elasticity indicating the important role of fibrinogen as a marker of vascular damage [18].

From the Figure 2, it was evident that the receiver operator characteristic curve (ROC) analysis demonstrated the area under the curve (AUC) of the preHTN subjects. The parameters Fibrinogen and MDH does not having the significant diagnostic ability to detect PreHTN. The hsCRP showed more AUC compared to the other markers (0.59), however it is insignificant. This might be due to limited sample size. The cut off levels of fibrinogen 366 mg/dl whereas malondialdehyde 3.5nmol/L and hsCRP greater than 1.1 mg/L has been considered as optimum cut off for predicting the pre HTN and to set a warning signal forprecautionary measures to be adopted immediately to in apparently normal healthy individual. Whereas it is evident from the Figure 3, all the three parameters showed significant AUC. The studies reported the ROC for hsCRP is highly sensitive [19].In the present study, none of the markers exhibit reliable diagnostic sensitivity and specificity in pre HTN. However, to some extent the CRP shown elevation in compared to other markers and in comparison to other markers hsCRP can be the independent predictive of long term HTN [22].Based on AUC, the cut off value has been determined for hsCRP to predict the pre HTN is 1.9 indicating an alarming level for the progression of pre HTN to stage I HTN (Table 2). Our study observed from Table 3, the diagnostic sensitivity is maximum and the cut off for these parameters i.e. of fibrinogen (425mg/dl), MDH (5.3nmol/ml) and hsCRP (2.75mg/l) indicating beyond this level the pre HTN may progress to stage II HTN. The ROC analysis of these markers in pre HTN showed insignificant area under the curve. This is because of very limited sample size.

Summary

- The present study was under taken to observe the levels of markers of pre and stage I HTN.
- Among the parameters, fibrinogen, MDH and hsCRP, the serum hsCRP have showed slight elevation the pre hypertension group compared to control and can be treated as predictive marker provided by analysing the more sample size. Based on AUC, the cutoff values of these markers has been established for fibrinogen, MDH and hsCRP helps in prediction and understanding the preHTN.
- The serum fibrinogen, MDH and hsCRP have shown an increase in the stage I hypertension group compared to control and pre HTN which are statistically significant fibrinogen (425 mg/dl), MDH (5.9 nmol/ml) and hsCRP (2.75mg/l). As per these findings, the cut off levels helps in predicting the stageI HTN and beyond these values understanding the progression to stage II HTN.

Conclusion

Nearly two-thirds of population are unaware of their pre and hypertensive conditions and the untimely diagnosis of prehypertension results in hypertension soon. The early and accurate diagnosis of pre and hypertension is still challenging and a mix up. There is paucity and diversified literature regarding markers in predicting the hypertension. Therefore, the biomarker hsCRP showed slight association in predicting the hypertension. This work tried toestablish cut off levels in predicting for pre hypertension and its progression.However, due to limited population of the study group it is still required further studies to establish optimum cut off to include as regular panel marker.

References

1. Michael J. Bloch, M.J. Worldwide prevalence of hypertension exceeds 1.3 billion Bloch. J. Am. Soc. Hypertens.2016; 10(10):753-754.
2. Amar S, Olanrewaju E, Hemant G. Circulating blood biomarkers in essential hypertension: a literature review. Can Fam Physician. 2008;54(10):1418-1423.
3. Parti G, Mutti E, Ravogli A, Villani A, Mancia G. Advantages and disadvantages of non-invasive ambulatory blood pressure monitoring. J Hypertens Suppl. 1990;8(6):33-38
4. Godwin M, Pike A, Kirby A, Jewer C, Murohy L. Prehypertension and hypertension in a primary care practice. Can Fam Physician. 2008;54(10):1418-23.
5. Svetkey L. Hypertension. 2005;(45):1056-1061.

DOI: 10.69605/ijlbpr_13.8.2024.71

6. Shere A, Eletta O, Goyal M. Circulating blood biomarkers in essential hypertension: a literature review. J Lab Precis Med. 2017; 2(12): 1-11.
7. Chobanian AV, Bakris GL, Black HR. "The Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure: the JNC 7 report". JAm Med Assoc. 2003; 289(19):2560-72.
8. Raghupathy A, Nanda K, Hira P, Hasan K. Hypertension in India:a systematic review and meta-analysis of prevalence, awareness,and control of hypertension,Int. J.Hypertens. 2014;32(6)
9. Sivasubramanian,RamakrishnanGeevar,Zachariah KartikGuptaJ,ShivkumarRao,P. P.Mohanan,K.Venugopal,SantoshSateesh,RishiSethi,DharmendraJain,NeilBardole I,KalaivaniMani Prevalence of hypertension among Indian adults: Results from the great India blood pressure survey pane. Heart J.2019; 4(71): 309-313
10. Reza J, Azadeh M, AliJ, Afsaneh R.A comparative study of the management of stage 2 hypertension by combined therapy with Losartan, Amlodipine and Hydrochlorothiazide. Int.Cardiovasc.Res.J.2012;6(3):79-83
11. Campbell NR, Lackland DT, Lisheng L, Niebylski ML, Nilsson PM, Zhang XH. "Using the Global Burden of Disease study to assist development of nation-specific fact sheets to promote prevention and control of hypertension andreductionindietarysalt:aresourcefromtheWorld HypertensionLeague". Journal of Clinical Hypertension. 17 (3): 165–7
12. Gbenga O, Thomas P. Principles and techniques of blood pressure measurement. CardiolClin. 2010; 28(4): 571–586.
13. Antonio, Ceriello, Possible Role of Oxidative Stress in the Pathogenesis of Hypertension. Diabetes Care 2008;(2):181-S184.
14. Raju, D. S. S. K., Munta, A. K., & Lalitha, D. L. (2021). A study on the serum arginase and nitrate levels in association with Fev1/Fvc ratio in chronic obstructive pulmonary disease.International Journal of Health Sciences,6(S9), 4803–4811.
15. Leite M. Uric acid and fibrinogen: age-modulated relationships with blood pressure components. Human Hypertension 2011;**(**25**):** 476–483
16. Tandon R,Sinha MK, Garg H, Khanna, H D Khanna. Oxidative stress in patients with essential hypertension. Natl Med J India. 2005;18(6):297-9.
17. D.S.S.K. Raju,D.L. Lalitha and P. Kiranmayi. A Study of Lipid Profile and Lipid Peroxidation in Chronic Kidney Disease with Special Reference to Hemodialysis. J Clinic Res Bioeth2013, 4:143.
18. Bray C, Bell LN, Liang H, Haykal R, Kaiksow F, Mazza JJ, Yale SH. "Erythrocyte SedimentationRate and C-reactive Protein Measurements and Their Relevance in Clinical Medicine". Wis. Med.J. (WMJ). 2016;**115**(6):317-21.
19. Wang T,Gona P, Martin G. Larson M, Levy D, Benjamin E,Tofler G, Jacques P, MeigsJ, Rifai N *et al*. Multiple Biomarkers and the Risk of Incident Hypertension.Hypertension.2007;(49):432-438.
20. Paul M Ridker, MPH; Nancy J. Brown, Douglas E. Vaughan, David G. Harrison, Jawahar L. Mehta, Established and Emerging Plasma Biomarkers in the Prediction of First Athero thrombotic Events. Circulation 2004;(109)
21. Hang C, Zhang J, Qin F *etal*. Evaluation of the predictive value of high sensitivity C reactive Protein in pregnancy induced hypertension syndrome.Exp. The Med. 2018;16 (2):619-622.
22. Conen, Zeller, Dieterle, B Martina.C-reactive protein and echocardiography have little impact on risk stratification in never-treated hypertensive patients. J Hum Hypertens. 2006;20(8):587-92.