

## Study Of Biological Markers Of Atherogenicity In Prehypertensives And Hypertensives: Association With Cardiovascular Risk

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**Abstract: Background & Objectives:** Prehypertension is a precursor of clinical hypertension and is closely related with the increased incidence of cardiovascular disease. Atherogenic lipid profile contributes to cardiovascular risk. We studied the prevalence of pre hypertension and its association with cardiovascular risk and biological markers of atherogenicity. **Method:** A total of 132 adults (age 30-50yrs) were enrolled. Based on the reported prevalence of prehypertension (38%) and hypertension (38%) in Madhya Pradesh, 50 prehypertensives and 50 hypertensives were selected for the study. Age and sex matched 50 normotensives served as control. Anthropometric measurements, fasting blood glucose and lipid variables were recorded. Lipid risk ratios were calculated and 10 years general cardiovascular risk was assessed by Framingham's heart scale. **Results:** High prevalence of cardiovascular risk factors (General obesity 60%,66%, Central obesity 60%,72%, Metabolic syndrome 39%,50%, Prediabetes 34%,44%,) in prehypertensives and hypertensives were noted. Prehypertensives having high normal blood pressure (130-139/85-89mmHg) carried higher cardiovascular risk (CVR  $7.88 \pm 7.17$ ) as compared to low prehypertensives (120-129/80-84mmHg, CVR  $3.22 \pm 2.38$ ). Hypertensives carried higher cardiovascular risk than prehypertensives. AIP was positively correlated with CVR. **Interpretation & Conclusion:** A high prevalence of prehypertension (66%) was observed. Major risk factors were male gender, obesity and metabolic syndrome. Atherogenic lipid profile in prehypertensives and hypertensives enhanced the cardiovascular risk.

**Key words:** AIP (Atherogenic index of plasma), cardiovascular risk, Prehypertension

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**Introduction:** The establishment of prehypertension category by the Seventh Joint National Committee on Prevention, Detection, Evaluation and Treatment of blood pressure, draws attention to a preclinical range of blood pressure (SBP 120-139 mmHg, DBP 80-89 mmHg) that has clinical and public health significance. Prehypertension is a risk factor for overt hypertension and increases cardiovascular risk. Prehypertension tends to cluster with other cardiovascular risk factors such as Obesity, dyslipidemia, insulin resistance, diabetes and metabolic syndrome<sup>1,2</sup>. The Atherogenic index of plasma (AIP) calculated as  $[\text{Log}(\text{TG}/\text{HDL-C})]$  provides information about the atherogenicity of plasma. AIP reflects delicate metabolic interaction within the whole lipoprotein complex<sup>3</sup>. Associated atherogenic lipoprotein phenotype with prehypertension considerably enhances the cardiovascular risk.

A high prevalence of prehypertension and hypertension exist in population of Madhya Pradesh<sup>4</sup>. We aim to study the 10 year general cardiovascular risk in prehypertensive and hypertensive subjects and its association with biological markers of atherogenicity and other cardiovascular risk factors in urban colony of Bhopal.

**Material:** The study was carried out in the Department of Physiology, Gandhi Medical College, and Bhopal (M.P.) in collaboration with the Department of Medicine of the institution, with the approval of protocol by the Ethics Committee of the faculty of Medicine, Gandhi Medical College, and Bhopal and after obtaining informed consent from the subjects. Community based cross sectional study was carried out. A total of 200 adults in the age range of 30-50 years were interviewed.

Based on the criteria scheduled in JNC-7<sup>5</sup>, 132 subjects were identified as prehypertensives and 68 as normotensives. Sample size was estimated on the basis of reported prevalence of prehypertension and hypertension in M.P. equal to 38%<sup>4</sup> Out of 132 prehypertensives and 68 normotensive, 50 normotensives and 50 prehypertensives, who fulfilled the inclusion criteria, were selected, including male (60%) and female (40%) for the study.

Hypertension was defined according to JNC-7 criteria<sup>5</sup>. Newly diagnosed subjects currently on antihypertensive medication were classified as hypertensives, irrespective of their current blood pressure readings. Age and Sex matched 50

hypertensive subjects, who fulfilled the inclusion criteria, were included in the study.

Each participant was given a validated questionnaire form to obtain their basic information which included age, sex, occupation, dietary intake, physical activity, smoking, personal history of hypertension, ischemic heart disease, diabetes mellitus and family history of hypertension, IHD and DM.

**Inclusion and Exclusion Criteria:**

**A. Normotensive Control Group-Inclusion Criteria**

- Healthy, asymptomatic subjects aged 30-50 years.
- Systolic blood pressure less than 120mmHg, diastolic blood pressure less than 80mmHg (JNC-7)
- Fasting blood glucose < 126mg/dl(ADA 2011).
- No ECG abnormality.
- No known endocrinal, metabolic, renal or cardiovascular disorder.

**Exclusion Criteria-**

- Age less than 30 years and more than 50 years.
- Fasting blood glucose  $\geq$  126mg/dl (ADA 2011).
- Subjects undergoing any kind of medical treatment.
- Subjects not willing to participate.

**B. Prehypertensive group-Inclusion Criteria-**

- Age 30-50 years.
- Systolic blood pressure of 120-139mmHg and/or diastolic blood pressure of 80-89mmHg (JNC-7).
- Fasting blood glucose < 126mg/dl (ADA 2011).
- No ECG abnormality.
- No known endocrinal, metabolic, renal or cardiovascular disorder.
- Subjects not undergoing any kind of medical treatment.

**Exclusion Criteria-**

- Age less than 30 years and more than 50 years.
- Baseline use of antihypertensive drugs, presence of normal blood pressure or hypertension (JNC-7).
- Fasting blood glucose  $\geq$  126mg/dl (ADA 2011).
- Systemic illness affecting cardiovascular system.

- Subjects undergoing any kind of medical treatment.
- Subjects not willing to participate.
- C. HYPERTENSIVE GROUP- Inclusion Criteria-
- Age 30-50 years.
- Diagnosed as hypertensive as per JNC-7 criteria systolic blood pressure  $\geq$  140mmHg and/or diastolic blood pressure  $\geq$  90mmHg by Physician, irrespective of the current blood pressure.
- Currently on antihypertensive treatment.
- Fasting blood glucose < 126mg/dl(ADA 2011)
- No ECG abnormality.
- No known endocrinal, metabolic and renal disorder.

**Exclusion Criteria-**

- Age less than 30years and more than 50 years.
- Prevalent cardiovascular disease (stroke, congestive heart failure, myocardial infarction or other manifestations of coronary heart disease.).
- Significant aortic and/or mitral valvular heart disease.
- Systemic illness affecting cardiovascular system.
- Fasting blood glucose  $\geq$  126mg/dl (ADA 2011).
- Subjects not willing to participate and those with acute illness.

**Method:** Brachial artery blood pressure (first and fifth Korotkoff Sounds) was measured three times consecutively with 15 minutes interval on seated participants after they had rested for 5 minutes, with the use of a standardized mercury Sphygmomanometer (Diamond, Industrial Electronic and Allied Products, Pune) and Stethoscope (3M Littman Classic II, German D). An appropriately sized cuff was placed on the right arm, pulse occlusion Pressure was determined, and the Cuff was inflated to 20 mmHg above that pressure. The mean of the last two of these measurements was used for estimation of blood pressure.

**Anthropometric Measurement:** Height was measured without shoes, in centimetre, using a standard stadiometer with subject standing in the erect posture. The readings were taken to the nearest 0.1 cm.

Weight was measured with the light clothes without shoes, in kilograms, using a professional calibrated weighing scale, rounded off to the nearest number. The BMI was calculated using the formula weight (kg)/height (m<sup>2</sup>).

Waist Circumference (WC) was measured at the midpoint between the lower border of ribcage and iliac crest with the subject standing erect with arms at the sides and feet placed about 25-30 cm apart with weight equally distributed on each leg. The circumference was measured in cm, to the nearest 0.5 cm at the end of normal expiration.

Biochemical Analysis: 5 ml of the fasting blood samples were collected and serum was separated for further analysis.

All analysis were measured in Auto analyzer (Merk 300) using Kits supplied by Agape Diagnostics, Kerala. Plasma glucose was measured by Glucose oxidase peroxidase technique<sup>6</sup>, serum total cholesterol and triglycerides were measured by CHOD-PAP method and GPO-PAP method<sup>7,8</sup>. Serum HDL-Cholesterol was measured after precipitation by Magnesium chloride phosphotungsted<sup>9</sup>. LDL-Cholesterol was calculated using friedewald's Formula (TC-(VLDL+HDL))<sup>10</sup>. Lipid risk ratios were calculated as TC/HDL-C, TG/HDL-C LDL-C/HDL-C, LDL-C/TC and atherogenic index of plasma (AIP) [Log (TG/HDL-C)]<sup>3</sup>.

Definition and Diagnostic Criteria: Overweight was defined as BMI  $\geq 23$  kg/m<sup>2</sup> and obesity as BMI of  $\geq 27.5$  kg/m<sup>2</sup>. While central obesity was defined as WC (cm) >90 in male and >80 in female.<sup>11,12</sup>

According to JNC -7 criteria, normal blood pressure was defined as systolic blood pressure <120 mmHg and diastolic blood pressure <80 mmHg and prehypertension as systolic blood pressure between 120 to 139 mmHg or diastolic blood pressure between 80 to 89 mmHg. Hypertension was defined as systolic blood pressure  $\geq 140$  mmHg or diastolic blood pressure  $\geq 90$  mmHg. All subjects currently on antihypertensive medications or having written prescriptions of antihypertensive drugs were classified as "hypertensive", irrespective of their current blood pressure readings.

Metabolic syndrome was defined according to the executive summary of the third report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation and treatment of high blood cholesterol in Adults (Adult Treatment Panel III)<sup>13</sup>

NCEP-ATP III Criteria required the presence of at least 3 of the following:

1. Abdominal obesity (cm) - male > 90, female > 80.
2. Dysglycemia fasting blood glucose (mg/dl) -  $\geq 100$ .
3. Hypertension (mmHg) -  $\geq 130/\geq 85$ .
4. High triglycerides (mg/dl) -  $\geq 150$ .
5. Low HDL – C (mg/dl) – male < 40, female < 50.

10 years general cardiovascular risk was assessed by using Framingham risk score based on a general cardiovascular risk profile for use in primary care : The Framingham Heart study (2008).<sup>14</sup>

Family history of hypertension and diabetes was defined by the presence of hypertension or diabetes in at least one parent of the enrolled subjects. subjects included in the study were screened for Pre diabetes and diabetes based on FBG (mg/dl) 100-125 mg/dl,  $\geq 126$  mg/dl as per American diabetes association (ADA 2011 criteria)<sup>15</sup>.

Statistical analysis: Statistical analysis was performed by SPSS 13.0 software. Results were expressed as mean  $\pm$  standard deviation. Student "t" test was used to compare the parameters between normotensive (NT) controls, prehypertensive and hypertensive subjects. Correlation analysis was done using Karl Pearson's method. A two tailed P value of less than 0.05 was considered as significant for all statistical tests.

**Results:** Demographic and behavioural characteristics of the study population as per blood pressure groups are shown in Table-1. It is observed that all the anthropometric parameters showed a gradual upward trend from normotensive to hypertensive group. Prehypertensive category had significant higher levels of BMI, WC and blood pressure values in comparison to normotensive category (p value < 0.05), similarly in hypertensive subjects mean values of all the parameters measured were

significantly high in comparison to prehypertensives. 34% of the subjects were identified as prediabetic. Prehypertensive subjects showed significantly higher frequency of family history of hypertension and diabetes than the normotensive subjects. 40% of the hypertensive subjects had positive family history of hypertension and 24% had positive family history of diabetes.

**Table 1: Demographic and behavioural characteristics of the study population by blood pressure groups (Mean±SD)**

Variables	Normotensives N=50	Prehypertensives N=50	Hypertensives N=50
Mean age (yrs)	38.34±1.37	38.30±2.56	40.01±1.64
Sex, Male/ Female	30/20	30/20	30/20
Body Mass Index (BMI, kg/m <sup>2</sup> )	Nil (21.99±1.37)	60%(26.02±2.56)*	66%(26.28±3.64) <sup>†</sup>
Waist Circumference (WC,cm)•	8%(86.2±5.3)	60%(91.44±9.52)* <sup>†</sup>	72%(96.92±5.85) <sup>†</sup>
HR (bpm)	81.14±7.51	82.56±7.61 <sup>†</sup>	86.56±5.32 <sup>†</sup>
SBP (mmHg)	117.12±9.28	133.14±10.17* <sup>†</sup>	144.52±7.45 <sup>†</sup>
DBP (mmHg)	76.52±5.81	87.26±6.14* <sup>†</sup>	92.52±4.62 <sup>†</sup>
PREDIABETICS	14%	34%	44%
FAMILY HISTORY -			
Hypertension	16%	32%	40%
Diabetes Mellitus	08%	18%	24%
Ischaemic Heart Disease	3%	12%	23%
BEHAVIOURAL CHARACTERISTICS-			
SMOKING	16%	18%	34%
SEDENTARY PHYSICAL ACTIVITY	22%	28%	36%
DIETARY	Non Veg -22% Veg-78%	Non Veg-28 Veg-72%	Non Veg-34% Veg-66%

\* P-value (<0.05) for comparison between normotensive and prehypertensive subjects. <sup>†</sup> P-value (<0.05) for comparison between prehypertensive and hypertensive subjects. • P-value (<0.05) for comparison between normotensive and hypertensive subjects.

Central Obesity (Indian Criteria) WC (cm)>90 in Men and in Women >80. •

Metabolic Syndrome ATP III Criteria: Metabolic characteristics of the study populations are shown in Table -2. It is observed that except for HDL-C all other biochemical parameters increased from normotensives to hypertensives including lipid risk ratios. In prehypertension group, subjects with metabolic syndrome exhibited higher values of lipid fractions except HDL-C as compared to subjects not having metabolic syndrome.

**Table 2: Metabolic characteristics of study population by blood pressure group (Mean±SD)**

Parameters	Normotensives N=50	Prehypertensives N=50	Hypertensives N=50
FBG (mg/dl)	76.83±3.32	80.21±2.81* <sup>†</sup>	85.20±3.82 <sup>†</sup>
TC (mg/dl)	147.46±28.25	178.28±22.98* <sup>†</sup>	254.14±52.79 <sup>†</sup>
TG (mg/dl)	113.02±30.42	159.51±21.86* <sup>†</sup>	194.28±20.42 <sup>†</sup>
LDL-C (mg/dl)	79.94±25.47	109.51±23.98* <sup>†</sup>	184.22±50 <sup>†</sup>
HDL-C (mg/dl)	46.48±11.04	34.92±6.43* <sup>†</sup>	31.22±2.46 <sup>†</sup>
TC/HDL-C	3.25±0.88	5.29±1.23* <sup>†</sup>	6.39±2.4 <sup>†</sup>
TG/HDL-C	2.53±0.82	4.76±1.32* <sup>†</sup>	4.48±1.49 <sup>†</sup>
LDL-C/HDL-C	1.80±0.72	3.27±1.02* <sup>†</sup>	4.59±2.05 <sup>†</sup>
LDL-C/TC	0.52±0.09	0.60±0.06* <sup>†</sup>	0.69±0.07 <sup>†</sup>
AIP	0.14±0.23	0.32±0.14* <sup>†</sup>	0.42±0.50 <sup>†</sup>

\* P-value (<0.05) for comparison between normotensive and prehypertensive subjects. <sup>†</sup> P-value (<0.05) for comparison between prehypertensive and hypertensive subjects. • P-value (<0.05) for comparison between normotensive and hypertensive subjects

**Table 3: Biochemical profile of subjects having metabolic syndrome**

Variables	Prehypertensives N=19	Hypertensives N=25
FBG (mg/dl)	80.21±2.81 <sup>†</sup>	85.20±3.82
TC (mg/dl)	182.87±22.53 <sup>†</sup>	254.14±52.79
TG (mg/dl)	167±18.39 <sup>†</sup>	194.28±20.42
LDL-C (mg/dl)	114.05±22.4 <sup>†</sup>	184.22±50
HDL-C (mg/dl)	33.05±5.68 <sup>†</sup>	31.22±2.46

TC/HDL-C	5.65±1.11	6.39±2.4
TG/HDL-C	5.17±1.18 <sup>†</sup>	4.48±1.49
LDL-C/HDL-C	3.54±0.95 <sup>†</sup>	6.39±2.4
LDL-C/TC	0.61±0.06 <sup>†</sup>	0.69±0.07
AIP	0.36±0.09	0.42±0.50

<sup>†</sup> P-value (0.05) for comparison between prehypertensive and hypertensive subjects with metabolic syndrome.

**Table 4: Cardiovascular risk (CVR) and atherogenic index of plasma (AIP) by blood pressure groups (Mean ±SD)**

CV RISK CATEGORY	VARIABLES	NORMOTENSIVE N=50	PREHYPER TENSIVE N=50	HYPER TENSIVE N=50
<10%	CVR	3.47±2.17	5.21±2.48* <sup>†</sup>	6.84±2.0 <sup>+</sup>
	AIP	0.12±0.15	0.28±0.15*	0.34±0.23 <sup>+</sup>
10-20%	CVR	10.13±2.54	12.13±1.31* <sup>†</sup>	14.1±2.64 <sup>+</sup>
	AIP	0.18±0.10	0.35±0.9*	0.35±0.05 <sup>+</sup>
>20%	CVR	-	28.07±4.31*	28.46±5.77 <sup>+</sup>
	AIP	-	0.41±0.04* <sup>†</sup>	0.43±0.05 <sup>+</sup>

Results of cardiovascular risk assessment and atherogenic index of plasma are displayed in Table -4. A significant increase in cardiovascular risk was observed for both prehypertensives and hypertensives as compared to normotensives. Prehypertensives with metabolic syndrome in greater than 20% cardiovascular risk category found exposed to high cardiovascular risk (27.19±5.02). Statistically significant increase in percent cardiovascular risk (p <0.05) was observed between all blood pressure groups. A rising trend of atherogenic index of plasma was observed from low risk category to high risk category in all blood pressure groups.

\* P-value (<0.05) for comparison between normotensive and prehypertensive subjects. <sup>†</sup> P-value (<0.05) for comparison between pre hypertensive and hypertensive subjects. <sup>+</sup> P-value (0.05) for comparison between normotensive and hypertensive subjects. • 10 yrs general cardiovascular risk (Framingham's Heart Scale) 14 • Atherogenic Index of Plasma [Log (TG/HDL-C)]<sup>3</sup> Prehypertensives having high normal blood pressure (130-139/85-89mmHg) carried higher cardiovascular risk (CVR 7.88±7.17) as compared to low prehypertensives (120-129/80-84mmHg, CVR 3.22±2.38). Hypertensives carried higher cardiovascular risk (26.73±5.42) then prehypertensives (6.56±6.48).

Atherogenic index of plasma (AIP) [log (TG/HDL-C)] showed significant positive correlation with BMI (r=0.31, 0.45) and 10 years general cardiovascular risk (r=0.27, 0.55) in prehypertensive with metabolic syndrome and hypertensive subjects.

**Discussion:** Report of JNC-7 acknowledges the continuous relationship between blood pressure and cardiovascular diseases. In the light of published data suggesting increase in prevalence of hypertension and prehypertension in India, Shantirani et al<sup>16</sup> (2003)-47%, Prabhakaran et al<sup>17</sup> (2005)-44%, Chockalingam A. et al<sup>18</sup> (2005)-42.5%, Hazarika NC. et al<sup>19</sup> (2004)-54%, the present study explores the prevalence of prehypertension and 10 years general cardiovascular risk, in an Indian middle socioeconomic population, residing in urban colony of Bhopal, Madhya Pradesh. The high prevalence of prehypertension in the current study (66%) confirms the increasing trend. Male showed higher percentage of prehypertension (60%) compared to females (40%), possibly due to increased prevalence of metabolic and cardiovascular risk factors, similar findings were reported by Asmathulla S. et al.<sup>20</sup> (2011).

In the current study, prehypertension was highest in age group 30-39yrs (52%) as compared to age group 40-50yrs (48%). The findings are in harmony with the findings reported by Yadav S. et al<sup>21</sup> (2008), Mohan V. et al<sup>22</sup> (2007).

There was a high prevalence of cardiovascular risk factors in the study population (General obesity : 60%, 66%, Central obesity: 60%, 72%, Smoking: 18%, 34%, Prediabetes: 34%, 44%) in prehypertensives and hypertensives respectively. Two or more cardiovascular risk factors were present in higher proportion of hypertensives and prehypertensives compared to normotensive subjects, similar findings were reported by Yadav S. et al (2008).<sup>21</sup>

In the current study, existence of metabolic syndrome was 39% in prehypertensives and 50% in hypertensives.

High normal prehypertension (130-139/85-89mmHg) was associated with increased 10 yrs general cardiovascular risk as compared to low prehypertension (120-129/80-84mmHg), similar

finding were reported by Heather A. et al<sup>23</sup>(2005).

Increasing trend of cardiovascular risk was observed in all blood pressure categories .Because our objective was to evaluate the association of atherogenic indices with prehypertension and hypertension, an attempt had been made to establish the correlation, if any.

Atherogenic index of plasma (AIP) [log (TG/HDL-C)] showed significant positive correlation with BMI and 10 years general cardiovascular risk in prehypertensive and hypertensive subjects. Berenson G.S. et al<sup>24</sup> (1998) reported that atherogenic indices had additive effect that contribute to the increase of atherosclerosis in arterial wall thickness.

Limitations of our study deserved to be mentioned, blood pressure measurement were taken on a single day, average of last two readings was taken. Prehypertension was defined based on the average of 3 readings of blood pressure. Use of 24-h blood pressure, ambulatory measurements as gold standard test for diagnosing the prehypertension and hypertension could not be used. Because of the small sample size it was not possible to infer with certainty whether AIP is positively associated with prehypertension or not.

The strength of our study includes the sample of adults from a colony which had representative mix of subjects from different communities.

**Interpretation and Conclusion:** A high prevalence of prehypertension was observed in urban adults of Bhopal Madhya Pradesh. Prehypertensives carried a greater cardiovascular risk as compared to normotensives. Atherogenic index of plasma showed significant positive association with cardiovascular risk in both prehypertensives and hypertensives. As the prevalence of prehypertension is increasing, effective preventive measures are to be implemented to reduce the prevalence of hypertension in Madhya Pradesh.

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