

# MATERIALS AND METHODS

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#### 1. MATERIALS.

#### A) STUDY SAMPLE

The cross-sectional, comparative study is taken from outdoor as well as indoor patients of hypertension at Shri Sayajirao General Hospital, Vadodara. Healthy matched controls with comparable baseline characteristics were taken from general population.

The study was conducted for a period of one year from May 2004 to April 2005

## B) <u>SELECTION OF STUDY SUBJECTS</u> INCLUSION CRITERIA

Cases:

Patients diagnosed as having hypertension (BP  $\geq$  140/90 mill) as defined by the JNC-7 guidelines.

Patients on life style modification or drug therapy for hypertension.

Controls:

Healthy non-hypertensive subjects (as proven by 2BP recordings on 2 separate visits) with comparable baseline characteristics.

## **EXCLUSION CRITIRIA**

- Presence of diabetes.
- History of cerebrovascular stroke.
- History of coronary artery disease.
- History of peripheral vascular disease.
- Presence of any acute illness or fever.
- S. Total cholesterol > 240 mg%
- S. LDL cholesterol > 160 mg%
- Overweight or Obesity Body mass index  $\geq 25 \text{ kg/m}^2$

### 2 <u>METHODS</u>

## A) History and Clinical examination

Each patient enrolled in the study was asked detailed history regarding hypertension and its complications.

All previous medical records were checked. History about diet, bladder & bowed habits, alcoholism and smoking was inquired.

During general examination, vital signs, and anthropometric measurements were taken. Search for any sign of atherosclerosis was made and all peripheral pulsations were checked. Examination of central nervous system including optic fundus, cardiovascular system, respiratory system and alimentary system was performed.

#### 3 **INVESTIGATIONS**

Blood was drawn for complete haemogram, blood sugars, blood urea, S.cretinine, S.lipid profile. Urine examination was done ECG and Chest x-ray were done

Endothelial function was studied in accordance with the 2002 American college of Cardiology (ACC) guidelines (5) for ultrasound assessment of endothelial-dependant Flow-mediated Dilation (FMD) of the brachial artery in the following manner:

#### SUBJECT PREPARATION

Abstinence from smoking, tobacco chewing, ingestion of alcohol and high fat foods for at least 8 hours prior to study was advised. All the vasoactive medications like aspirin or other anti-inflammatory drugs if present stopped at least 2 days prior. Study in period of active menstrual bleeding was avoided.

Study was performed in a quiet, temperature controlled room.

#### **RESTING BRACHIAL ARTERY DIAMETER**

The subject is positioned with the arm in a comfortable position.

The brachial artery is imaged above the antecubital fossa in a longitudinal plane and the internal diameter is noted using a high quality ultrasound Doppler probe.

This is the resting brachial artery is diameter. (a)

# ENDOTHELIAL DEPENDANT DILATATION / FLOW - MEDIATED DILATATION (FMD)

After measuring the resting brachial artery diameter, a sphygmomanometric cuff is secured above the antecubital fossa and is inflated 50 mm above systolic pressure for 4 minutes.

This creates a flow stimulus and the brachial artery dilates in response to this. This internal lumen diameter of the brachial artery is measured at 60 seconds after deflating the cuff. This is the post-sheer stress diameter. (b)

The percentage of this change of diameter from the resting diameter is taken as the flow mediated dilatation (FMD %)

FMD% = 
$$\frac{(Post sheer stress - Resting Diameter)}{Resting Diameter (a)} X 100$$

This is the endothelium dependant brachial artery dilatation as it is effected by endogenous vasodilator mediators released by the endothelium of the artery in response to stress.

#### **ENDOTHELIUM INDEPENDENT DILATATION:**

After this, a period of at least 10 minutes of rest is given to the subject. Then a single dose of 400 micrograms of glyceryl trinitrate (GTN) tablet is given sublingually. The diameter of the brachial artery is noted 4 minutes after giving the nitrate. (c)

The percentage change from the resting diameter is taken as glyceryl trinitrate (GTN).

GTN % = <u>Ie Post GTN diameter – Baseline diameter</u> X 100 Baseline diameter (a) This is the endothelium – independent dilatation as it is effected by the exogenous nitrate and the vasodilator mediators produced by the endothelium of the brachial artery play no role in this effect.