“A STUDY TO EVALUATE THE ASSOCIATION OF BIOMARKERS WITH PULMONARY CAPILLARY WEDGE PRESSURE AND PLASMA CONCENTRATION OF THE INOTROPE DOBUTAMINE IN PATIENTS UNDERGOING ON-PUMP CABG SURGERY IN A SUPER-SPECIALITY HOSPITAL AT COIMBATORE : A PROSPECTIVE OBSERVATIONAL STUDY”

DISSERTATION

SUBMITTED FOR

THE TAMILNADU DR. M.G.R. MEDICAL UNIVERSITY

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DEPARTMENT OF PHARMACOLOGY

PSG INSTITUTE OF MEDICAL SCIENCES AND RESEARCH

PEELAMEDU, COIMBATORE – 641004

TAMILNADU, INDIA

MAY 2020

PSG INSTITUTE OF MEDICAL SCIENCES & RESEARCH COIMBATORE

CERTIFICATE-I

This is to certify that this dissertation entitled **"A Study To Evaluate The Association Of Biomarkers With Pulmonary Capillary Wedge Pressure And Plasma Concentration Of The Inotrope Dobutamine In Patients Undergoing On-Pump CABG Surgery In A Super-Speciality Hospital At Coimbatore : A Prospective Observational Study",** is an original work done by **Dr.R.SUBHASHINI**, Postgraduate under the guidance of **Dr.K.BHUVANESWARI**, M.D.,PGDBE., Professor and Head, Department of Pharmacology, **PSG IMS&R.**

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DECLARATION BY THE CANDIDATE

I hereby declare that this dissertation entitled, **"A Study To Evaluate The Association Of Biomarkers With Pulmonary Capillary Wedge Pressure And Plasma Concentration Of The Inotrope Dobutamine In Patients Undergoing On-Pump CABG Surgery In A Super-Speciality Hospital At Coimbatore : A Prospective Observational Study",** is a bonafide work done by me under the guidance and supervision of **Dr.K.Bhuvaneswari**, Professor & HOD, Department of Pharmacology, PSG Institute of Medical Sciences & Research. This study was conducted at the PSG Institute of Medical Sciences & Research, Coimbatore, under the aegis of The TamilnaduDr.MGR Medical University, Chennai, as part of the requirement for the award of M.D. Degree in Pharmacology.

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To
Dr R Subhashini
Postgraduate
Department of Pharmacology
Guide/s: Dr K Bhuvaneswari / Dr P R Murugesan
PSG IMS & R
Coimbatore

Ref: Project No. **17/370**

Date: December 27, 2017

Dear Dr Subhashini,

Institutional Human Ethics Committee, PSG IMS&R reviewed and discussed your application dated 05.12.2017 to conduct the research study entitled *"A study to evaluate the association of biomarkers with pulmonary capillary wedge pressure and plasma concentration of the Inotrope Dobutamine in patients undergoing on-pump CABG surgery in a super-specialty hospital at Coimbatore: A prospective observational study"* during the IHEC meeting held on 22.12.2017.

.The following documents were reviewed and approved:

1. Project submission form
2. Study protocol (Version 1 dated 05.12.2017)
3. Informed consent forms (Version 1 dated 05.12.2017)
4. Data collection tool (Version 1 dated 05.12.2017)
5. Permission letter from concerned Head of the Department
6. Current CVs of Principal investigator, Co-investigator
7. Budget

The following members of the Institutional Human Ethics Committee (IHEC) were present at the meeting held on 22.12.2017 at IHEC Secretariat, PSG IMS & R between 10.00 am and 11.00 am:

Sl. No.	Name of the Member of IHEC	Qualification	Area of Expertise	Gender	Affiliation to the Institution Yes/No	Present at the meeting Yes/No
1	Mr R Nandakumar (Chairperson, IHEC)	BA., BL	Legal Expert	Male	No	Yes
2	Dr D Vijaya (Member – Secretary, IHEC)	M Sc., Ph D	Basic Medical Sciences (Biochemistry)	Female	Yes	Yes
3	Dr S Shanthakumari	MD	Pathology, Ethicist	Female	Yes	Yes
4	Dr Sudha Ramalingam	MD	Epidemiologist, Ethicist Alt. member-Secretary	Female	Yes	Yes
5	Dr G Subhashini	MD	Epidemiologist	Female	Yes	Yes

The study is approved in its presented form. The decision was arrived at through consensus. Neither PI nor any of proposed study team members were present during the decision making of the IHEC. The IHEC functions

27/12

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in accordance with the ICH-GCP/ICMR/Schedule Y guidelines. The approval is valid until one year from the date of sanction. You may make a written request for renewal / extension of the validity, along with the submission of status report as decided by the IHEC.

Following points must be noted:

1. IHEC should be informed of the date of initiation of the study
2. Status report of the study should be submitted to the IHEC every 12 months
3. PI and other investigators should co-operate fully with IHEC, who will monitor the trial from time to time
4. At the time of PI's retirement/intention to leave the institute, study responsibility should be transferred to a colleague after obtaining clearance from HOD, Status report, including accounts details should be submitted to IHEC and extramural sponsors
5. In case of any new information or any SAE, which could affect any study, must be informed to IHEC and sponsors. The PI should report SAEs occurred for IHEC approved studies within 7 days of the occurrence of the SAE. If the SAE is 'Death', the IHEC Secretariat will receive the SAE reporting form within 24 hours of the occurrence
6. In the event of any protocol amendments, IHEC must be informed and the amendments should be highlighted in clear terms as follows:
   a. The exact alteration/amendment should be specified and indicated where the amendment occurred in the original project. (Page no. Clause no. etc.)
   b. Alteration in the budgetary status should be clearly indicated and the revised budget form should be submitted
   c. If the amendments require a change in the consent form, the copy of revised Consent Form should be submitted to Ethics Committee for approval
   d. If the amendment demands a re-look at the toxicity or side effects to patients, the same should be documented
   e. If there are any amendments in the trial design, these must be incorporated in the protocol, and other study documents. These revised documents should be submitted for approval of the IHEC and only then can they be implemented
   f. Any deviation-Violation/waiver in the protocol must be informed to the IHEC within the stipulated period for review
7. Final report along with summary of findings and presentations/publications if any on closure of the study should be submitted to IHEC

Kindly note this approval is subject to ratification in the forthcoming full board review meeting of the IHEC.

Thanking You,

Yours Sincerely,

27/12/17

SECRETARY
PSG IMSR
COIMBATORE 641004

Dr Sudha Ramalingam
Alternate Member - Secretary
Institutional Human Ethics Committee

Clinical Trial Details (PDF Generation Date :- Wed, 06 Nov 2019 11:37:54 GMT)

CTRI Number	CTRI/2018/05/013554 [Registered on: 01/05/2018] - **Trial Registered Prospectively**
Last Modified On	27/04/2018
Post Graduate Thesis	Yes
Type of Trial	Observational
Type of Study	Cross Sectional Study
Study Design	Other
Public Title of Study	An Observational Study To Relate Two Biological markers with Left Heart Pressure And Blood Concentration of A Drug Dobutamine In Patients Undergoing By Pass Heart Surgery
Scientific Title of Study	A Study To Evaluate The Association Of Biomarkers With Pulmonary Capillary Wedge Pressure And Plasma Concentration Of The Inotrope Dobutamine In Patients Undergoing On-Pump CABG Surgery In A Super-Speciality Hospital At Coimbatore : A Prospective Observational Study

Secondary IDs if Any

Secondary ID	Identifier
NIL	NIL

Details of Principal Investigator or overall Trial Coordinator (multi-center study)

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CERTIFICATE – II

This is to certify that this dissertation work titled **"A Study To Evaluate The Association Of Biomarkers With Pulmonary Capillary Wedge Pressure And Plasma Concentration Of The Inotrope Dobutamine In Patients Undergoing On-Pump CABG Surgery In A Super-Speciality Hospital At Coimbatore : A Prospective Observational Study",** of the candidate Dr.R.Subhashini with registration Number **201716302** for the award of M.D. Degree in the branch of Pharmacology.

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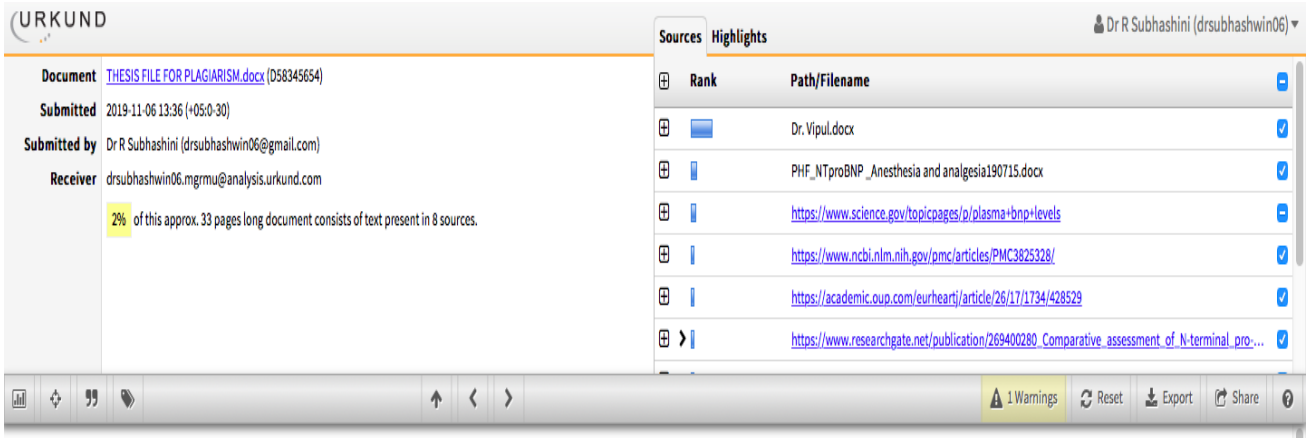

ABSTRACT Background: On-Pump Coronary Artery Bypass Grafting (CABG) is considered the gold standard surgical revascularization procedure. On-Pump CABG surgery uses a Cardio-Pulmonary Bypass machine that enables the entire cardiac output to circumvent the patient's heart and lungs. The intra-operative Pulmonary Capillary Wedge Pressure (PCWP) reflects Left ventricular End Diastolic Volume, which is increased in case of left ventricular systolic dysfunction. Such a dysfunction demands judicious use of inotropes to improve myocardial contractility. The cardiac biomarker NT-Pro BNP is an acknowledged marker of Left Ventricular Systolic dysfunction that results in Low Cardiac Output Syndrome (LCOS). The inotrope that is commonly used to tackle a LCOS is Dobutamine. Cardio Pulmonary Bypass machine employed during an On-Pump CABG surgery elicits a Systemic Inflammatory Response Syndrome (SIRS) with the production of various inflammatory cytokines which are known to influence the pharmacokinetics of various drugs administered during the course of the procedure. Aim: To investigate the role of a cardiac biomarker NT-Pro BNP and an inflammatory marker TNF-α on the Pulmonary Capillary Wedge Pressure (PCWP) and the plasma concentration of the inotrope dobutamine in patients undergoing On-Pump CABG surgery.

Primary Objective: To evaluate if the preoperative level of the cardiac biomarker NT-Pro BNP, correlates with intraoperatively measured Pulmonary Capillary Wedge Pressure (PCWP) in patients undergoing On-Pump CABG surgery. Secondary Objective: To evaluate the role of the inflammatory mediator TNF alpha on the intraoperative plasma concentration of Dobutamine. Tertiary Objective: To evaluate the association of NT-pro BNP and TNF-α with existing comorbidities. Materials and Method: Between July 2018 and June 2019 , 44 patients who underwent On-

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I express my deepest gratitude and sincere thanks to my beloved teacher **Dr.K.Bhuvaneswari M.D., PGDBE**, Professor and Head , Department of Pharmacology of PSG Institute of Medical Sciences & Research, for being my guide. It was her mentoring, valuable suggestions, guidance and constant encouragement in every step that has helped me to complete my research work.

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TABLE OF CONTENTS

INTRODUCTION

Cardiovascular Disease (CVD) burden is on the rise globally and is recognized to be the leading cause of mortality. According to the WHO, of the 41 million deaths due to Non Communicable Diseases (NCDs) worldwide, Cardiovascular Disease claims lives of 17.9million people every year, accounting for 44% of all deaths due to NCDs. [1] In India, CVD death rates have escalated from 155.7 to 209.1 per 100,000 persons between 1990 and 2016. [2]

In the United States of America, Coronary Heart Disease (CHD) was found to be the prime cause (43.2%) of mortalitydue to CVD. By 2030 medical expenditure due to Coronary Heart Disease is projected to increase by about 100%. [3] Similarly in India majority of deaths due to CVD are attributed to Coronary Artery Disease (CAD) followed by stroke, both of which account for 83% of deaths due to CVD. [4]

Stable Coronary Artery Disease is characterized by a history of Myocardial Infarction (MI) or identification of coronary atherosclerotic plaque by catheterization or computed tomography angiography in the absence of acute coronarythrombosis. As long as patients are asymptomatic or their symptoms are kept under control by medications or revascularization they are considered stable. [5]

However in conditions where pharmacotherapy or Percutaneous Coronary Intervention (PCI) turns out to be unsuccessful as in cases such as advanced Ischemic Heart Disease, when Left main coronary artery stenosis is $\geq 50\%$, presence of Triple Vessel Disease (TVD) with or without Left Anterior Descending (LAD) artery disease, diagnosis of abnormal Left Ventricular Function (LVF) characterized by Ejection Fraction(EF)between 35% to 50%, surgical revascularisation by Coronary

Artery Bypass Grafting (CABG) is considered the gold standard modality of management. [5,6]

In CABG surgery a blood vessel section from the aorta is grafted to the coronary artery to bypass the occluded section of the coronary artery and improve the blood flow to the heart. It may be performed either On-Pump or Off-Pump. In On-Pump CABG surgery a combination ofCardioplegia and Cardio-Pulmonary Bypass (CPB) machine enables the entire cardiac output to circumvent the patient's heart and lungs during the course of grafting.[7]On Pump CABG requires the use of inotropes during the course of the procedure, while weaning the patient of the pump and in the perioperative period for ensuring adequate cardiac contractility resulting insufficient cardiac output.

Dobutamine is approved by the Food and Drug Administration (FDA) for short-term use in conditions associated with reduced contractility due to heart failure or cardiac decompensation as a result of cardiac surgical procedures.Dobutamine is a synthetic sympathomimetic amine withprominent β-1 adrenergic agonist properties which produces positive inotropic actionon the myocardium resulting in reduced End Systolic Volume (ESV) and increased Cardiac Output. This causes the baroreceptor mediated response to reduce the peripheral vascular resistance hence there is little or no change in the arterial blood pressure In addition dobutamine has also been found to have some β – 2 adrenergic agonistic activity, which contributes to the reduction in the systemic vascular resistance, and to a lesser extent has α-1 vasoconstrictor activity. However these effects are negated by the baroreceptor mediated response and beta 2 activity.[8]

B-type natriuretic peptide (BNP) is a peptide hormoneprimarily synthesized and secreted by cardiac myocytes of the left ventricle in response to ventricular stressas a result of pressure or volume expansion of the ventricle.BNP serves to maintain cardio-renal homeostasis.[8,9]

BNP is initially produced as a pre prohormone (pre pro BNP) which gets cleaved immediately after release to a BNP Precursor (proBNP).Pro-BNP undergoesfurther physiological cleavage into a biologically active BNP (C-terminal region) and an inactive N-terminal proBNP (NT-proBNP). Biologically active BNP and NT-pro BNP are hence secreted in a 1:1 ratio. However , NT-Pro BNP is found to have a half life of around 120 minutes in comparison to BNP whose half life is only20 minutes .This results in six fold higher levels of NT-Pro BNP than BNP although both are secreted in equimolar concentrations. BNP and NT-proBNP are cardiac biomarkers that are currently powerful diagnostic tools for detecting the presence and severity of Acute Heart Failure (AHF). They also serve as prognostic tools to determine the response to treatment for cardiac dysfunction and guide further therapy.In addition they are also screening tools that detect Left ventricular systolic and diastolic dysfunction .Among BNP and NT-Pro BNP, the latter is preferred as an investigational tool due to its longer half-life , higher levels in plasma , better stability at room temperature and lesser physiological variation.[10-15]The cardiac biomarker NT-Pro BNP is an acknowledged marker of Left Ventricular Systolic dysfunction which causes Low Cardiac Output Syndrome (LCOS).[11]

Pulmonary Capillary Wedge Pressure (PCWP) is an indirect predictor of Left Atrial Pressure (LAP). It is measured during CABG surgery using a balloon-tipped, multi-lumen catheter (Swan-Ganz catheter).In the absence of mitral stenosis the Left Atrium, Left Ventricle and the Pulmonary Venous system form a continuous circuit, hence in such circumstances the PCWP reflects Left Ventricular End-Diastolic Pressure (LVEDP) which is increased in case of Left Ventricular Systolic Dysfunction.[16] While weaning the patient off the CPB machine and in the post-operative period due to inadequate myocardial contractility patient is prone to develop LCOS characterised by raised LVEDP and as a result raised PCWP.

A good correlation of NT-Pro BNP with PCWP has been reported when patients were recompensated following decompensated heart failure.[21]

The CPB machine employed during the course of On-Pump CABG surgery results in complex activation of cellular and humoral inflammatory mediators such as TNF-alpha, IL-6, IL-8 , coagulation and complement cascades resulting in Systemic Inflammatory Response Syndrome (SIRS).Herein inflammatory response spreads throught the systemic circulation influence the pharmacokinetics of various drugs administered during the course of the procedure and postoperatively.[19,20]

In this study we hypothesize that , in a setting of On-Pump CABG surgery the cardiac biomarker NT-Pro BNP measured preoperatively would correlatewith the Pulmonary Capillary Wedge Pressure (PCWP) of patients measured intraoperatively. If acorrelation exits it would help to identify patients who are prone for LCOS during

surgery hence enabling risk stratification of patients prior to surgery. We also hypothesize that the concentration of the inotrope Dobutamine used to combat the LCOS is influenced by TNF-alpha released as a part of the SIRS elicited by the CPB machine. Such an influence on concentration of Dobutamine if exists, would necessitate higher dosing strategies with frequent dose titration.

AIM AND OBJECTIVES:

Aim of the Study:

To investigate the role of a cardiac biomarker NT-Pro BNP and an inflammatory marker TNF-α on the Pulmonary Capillary Wedge Pressure (PCWP) and the plasma concentration of the inotrope dobutamine in patients undergoing On-Pump CABG surgery.

Objectives:

Primary Objective: To evaluate if the preoperative level of the cardiac biomarker NT-Pro BNP, correlates with intraoperatively measured Pulmonary Capillary Wedge Pressure (PCWP) in patients undergoing On-Pump CABG surgery.

Secondary Objective: To evaluate the role of the inflammatory mediator TNF alpha on the intraoperative plasma concentration of Dobutamine.

Tertiary Objective: To evaluate the association of NT-pro BNP and TNF-α with existing comorbidities.

REVIEW OF LITERATURE

Coronary Artery Disease

Coronary Artery Disease (CAD) is one of the important causes of morbidity and mortality in bothdeveloping and developed countries.[22] The pathophysiological basis for CAD is the formation of atherosclerotic plaque(s) within the coronary vessels. Such atherosclerotic plaque(s)result in lumen obstruction andbecomes hemodynamically relevant once the extent of blockade exceeds 70%.[23]The plaque formed on the inner aspect of the coronary vessel can occlude blood flow through the vessel leading to ischemia or it can rupture following which, by the process of thrombus mediated occlusion of the vessel leads to the development of Acute Myocardial Infarction (AMI). Either of the two cases increases the risk of developing heart failure and/or death. The clinical presentation of CAD is in the form of ischemic chest pain (classically angina pectoris) with or without dyspnoea, the underlying mechanism of which is ischemic pump failure.[24]

Stable coronary artery disease is characterised byreversible nature of the demand-supply mismatch leading to ischemia, a Myocardial Infarction (MI) history or documentation of plaque by means of catheterization or CT angiography. When the condition isasymptomatic or the symptoms are managed effectively by pharmacotherapy or revascularization procedure, patients are considered stable.[25]

The aim of treatment of CAD is to provide symptomatic (abort or terminate) and prophylactic management of angina and also to prevent an event of Acute Myocardial

Infarction or Premature death.[26]Hence management strategies include lifestyle and risk factor modifications, antiplatelet and antianginal therapy. In spiteof an adequate trial of such pharmacotherapy when anginal symptoms are uncontrolled and plaque continues to progress , coronary revascularization with Percutaneous Coronary Intervention (PCI) or Coronary Artery Bypass Grafting (CABG) proves to be effective.[27-29]

Coronary Artery Bypass Grafting

Introduced in the mid-1960s, Coronary-artery bypass grafting (CABG) is still considered the gold standard for patients with extensive Coronary Artery Disease in whom PCI also fails to be an effective treatment option. Following its advent and subsequent improvisations, this procedure has contributed significantly to the reduction in deaths due to coronary artery disease in the last 5 decades.[31]

Coronary-artery bypass grafting (CABG) is a surgical coronary revascularization procedure wherein autologous arteries or veins serve as grafts in order to bypass partial or complete block of coronary arteries caused by atherosclerotic plaques.[30]

In the United States of America, CABG is one amongst the most common major surgical procedures and accounts for 4 ,00,000 operations performed per annum in the country.[32]

Indications for CABG Surgery includes: [28,29]

- Symptoms not amenable to Medical management / PCI
- Left Main Coronary Artery Stenosis (≥ 50%)
- Three-vessel disease with or without LAD artery disease
- Left ventricular dysfunction (Ejection Fraction < 35% to 50%)

On-Pump CABG Surgery

CABG is performed by means of a median (midline) sternotomy for adequate exposure. During the entire procedure muscles are not divided, and at the end of the procedure, through wire fixation the sternum is restored.[30]

Traditionally in order to permit scrupulousness to perform a successful CABG surgery, the heart is arrested. This is performed by cross clamping the ascending aorta which disconnects the heart from the systemic circulation. Following which the heart is perfused with cold, high-potassium cardioplegic solution that preserves myocardium by inhibition of the depolarization/repolarization cycle of myocytes.The function of the heart and lungs is taken over by a Cardiopulmonary-Bypass Machine, which offers perfusion pressure and oxygenation in order to maintain the circulation when an ischemic cardiac arrest is induced for a period of 1 to 2 hours.[30,35]

The bypass conduits that are frequently used are the Left Internal Thoracic Artery also known as Left Internal Mammary Artery (LIMA) and the Greater Saphenous Vein. Grafts from Saphenous veins are derived through small incisions in the thigh using endoscopic guidance.Such coronary arteries with proximal stenoses and patent distal vessels that are clinically significant as well are chosen for grafting. Caudal to the site

of stenosis an incision made in the coronary artery and the bypass graft is anastomosed end-to-side to the incision. The anastomosis procedure is enabled by optical magnification. Proximal anastomosis of the graft is finished by anastomosing the graft end-to-side to an aortotomy in the proximal ascending aorta however in in- situ arterial grafts (e.g., a left-internal- thoracic-artery graft) the arterial inflow is preserved. [33]

The duration of CABG surgery is typically around 3 to 4 hours. Post-surgery patients need to be hospitalized for 5 to 7 days. It takes a period of 6 to 12 weeks following discharge for complete recovery.[34]

Evidence Based Survival Benefit Of CABG Over PCI In Multivessel Disease

The Synergy between PCI with Taxus and Cardiac Surgery (SYNTAX) Trial randomized 1800 patients with triple vessel or left main coronary artery disease to either CABG or PCI.Individual participant assessment was done using SYNTAX score (measure of the extent and severity of coronary artery disease).SYNTAX scores classify severity of coronary artery disease as low (≤22), intermediate (23 to 32), or high (≥33).After 5 years of follow up those allocated to CABG, as compared to those allocated to PCI, had reduced rate of composite end point of death, myocardial infarction, stroke, or repeat revascularization ($P<0.001$).[36]

A meta-analysis of six Randomized Controlled Trials (RCT) comparing CABG with PCI in patients with multivessel disease found that for patients with complex multivessel disease and Diabetes Mellitusthe preferable procedure of revascularization

is CABG as it caused significant reductions in all-cause mortality (Number Needed to Treat (NNT) = 37 over four years) and MI (NNT = 26 over four years) .[37]

Low Cardiac Output Syndrome(LCOS) – Most Serious ComplicationOf CABG

Although CABG is a well-established procedurefor enhancing perfusion of the myocardium, it is associated with several complications mainlydue to the extracorporeal blood circulation. Among the common postoperative problems are dramatic hemodynamic changes, the most common and serious of which is LCOS.[40]

Low Cardiac Output Syndrome (LCOS) is an acute circulatory disorder associated with a prevalence of 2% to 10% in patients undergoing cardiac surgery.LCOS is defined by reduction in cardiac index to < 2.0 L/min/m^2, fall in systolic blood pressure to < 90 mmHg with signs of tissue hypoperfusion.On-pump CABG, emergency surgery and cardiopulmonary bypass (CPB) are independent significant risk factors.[45]

Pre-operatively whenLeft Ventricular Ejection Fraction (LVEF) is abnormal,there occurs a surge in prevalence of LCOS by 20%. In patients with CAD, LCOS causes further deterioration of the oxygen demand-supply mismatch that is further accentuated by CABG resulting in a very high mortality rate.[38,46]

The various contributors to LCOS after CABG are myocardial injury in the peri-operative period, pre-operative left ventricular systolic dysfunction (LVSD) and reperfusion injury induced myocardial stunning while weaning the patient off the CPB machine. In the post-operative period myocardial stunning is characterised by

abnormally low cardiac output necessitating positive inotropic agents for an uneventful surgery.[39]

Disconnection from the cardio pulmonary bypass machine must result in re-establishment of the circulation to the heart and lungs normally. The ability of the left and right ventricle to maintain the entire cardiac output decides the duration of the provisional period of partial bypass. Left ventricular dysfunction necessitates a period of partial bypass, simultaneously the state of ventricular loading is carefully manipulated by altering venous return and vascular resistance while myocardial contractility is enhanced by judicious use of positive inotropic agents. Hemodynamics and ventricular activity are determined by visual inspection of the heart and by Transoesophageal echocardiography (TEE). Hemodynamic management is aimed at maintaining four important parameters of cardiac function namely Heart rate and rhythm, Arterial pressure, Preload ventricular volume (Ventricular Filling pressure), and Contractility (Stroke volume).The clinical management goal following separation from CPB is to achieve aSystolic arterial pressure of 90-110 mm Hg, Cardiac index (CI) >2.0 L/min/m^2, a normal or low ventricular preload with filling pressure of 10-15 mmHg.[41]

LCOS complicating CABG causes increased mortality, and higher rates of morbidity manifesting as pulmonary complications, myocardial infarction, stroke, renal failure, and need for re surgery.[42-44] Also patients developing LCOS requirelonger duration of ventilatory support and Intensive Care Unit (ICU) and hospital stays, these by themselves further add as risk factors leading to still higher mortality. Longer the

ventilatory support, ICU and hospital stay, larger is the economic burden. Hence, it becomes vital to detect the predictors of LCOS following CABG surgery that would help in planning optimal strategiesto combat perioperative risk factors involved and ultimately reduce the incidence of LCOS following CABG.[45]

A multitude of treatment strategies are necessary to combat LCOS. It comprises of ventricular preload optimization prior to restoring systolic activity of the heart using positive inotropic agents with or withoutvasopressor therapy to re-establish adequate systemic vascular tone in states of vasoparesisor reduction inventricular after load when vascular resistance is increased. The ideal timing for treating LCOS is before the onset of ischemia induced end-organ injury culminating in organ failure.[47]

Among the various treatment options, the one of prime importance is the use of positive inotropic agents or mechanical circulatory support to improve myocardialcontractility and hemodynamic status subsequently.[48]Although many inotropic agents have been developed and evaluated dobutamine still remains the inotropic agent of choice for LCOS that develops in the perioperative period of CABG.[49]

DOBUTAMINE – Positive Inotropic Support

Introduced in the late 1970s Dobutamine, is a synthetic, intravenously administered catecholamine that has direct agonisticaction on β1 receptors predominantly and weak agonistic action on β2 and α1adrenergic receptors. At clinically used doses dobutamine exerts cardio selective action. Dobutamine is indicated for short term management of low cardiac output after cardiopulmonary bypass. Dobutamine is

about four times as potent as dopamine in increasing myocardial contractility at low concentrations.[50]Dobutamine is found to be superior to dopamine in the management of patients who are hemodynamically compromisedfollowing cardiac surgery, also in decreasing cardiac filling pressures and Peripheral Vascular Resistance (PVR) with comparatively less tachycardia.

At doses of 2.5 to 15μg/kg/min administered by continuous intravenous infusion , dobutamine enhances myocardial contractility improving cardiac output in LCOS accompanied by a slight increase in heart rate and a decrease in systemic vascular resistanceand PCWP. At higher doses vasoconstriction is observed.[51]

Dobutamine Chemistry:

Structurally Dobutamine resembles Dopamine but contains a bulky aromatic substituent at the amino group(Figure 1).Dobutamine is a racemic mixture (1:1) of positive and negative enantiomers. It is the positive isomer that is predominantly responsible for the potent β1and β2adrenoceptor agonist activity of dobutamine with minimal α1 adrenoceptor effects whereas the negative isomer has potent α1 adrenoceptor agonist with much weaker β1adrenoceptor agonistic activity and minimal β2adrenoceptor agonistic effect. Due to comparable adrenoceptor affinity of both the isomers, at higher concentrations the positive isomer inherently acts as a competitive inhibitor of the negative isomer.[53]

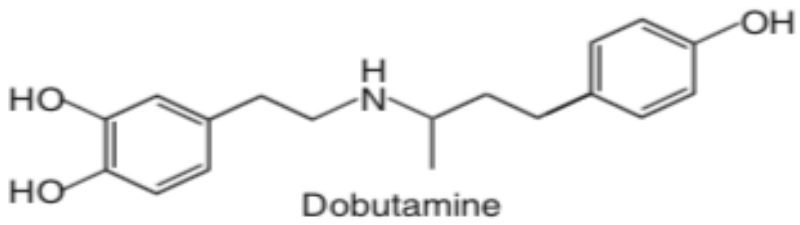


Fig 1: Molecular Structure of Dobutamine -Bulky Aromatic Substituent at Amino Group [52]

Dobutamine -Mechanism of Action[50]:

The Inotropes that help to stabilize LCOS share the common pathway of raising intracellular cyclic Adenosine MonoPhosphate (cAMP) resulting in increase in intracellular calcium concentrations. These include β-adrenergic agonists, endogenous catecholamines, and phosphodiesterase inhibitors.

The positive enantiomeric component of Dobutamine predominantly contributes to its action on β-1receptors. The guanine nucleotide regulatory cascade is activated via G proteins (Figure 2). β-1 receptors by their virtue of being coupled to G-proteins with G_sαsubunit leads to stimulation of the enzyme Adenylate Cyclase hence enhancing its activity. This results in conversion of Adenosine Triphosphate (ATP) to the intracellular second messenger cAMP. Intracellular cAMP stimulates release of calcium from the sarcoplasmic reticulum. The released calcium is used by contractile proteins and increasesmyocardial force of contraction raising stroke volume and cardiac output .In the vasculature, the α1-adrenergic agonist effect of the negative enantiomer is overcome by the partial agonistic activity of the positive enantiomer and also by the vasodilatory action caused by β_2-receptor stimulation. This usually results

in a moderate decrease in peripheral vascular resistances and venous filling pressures.[51,54]

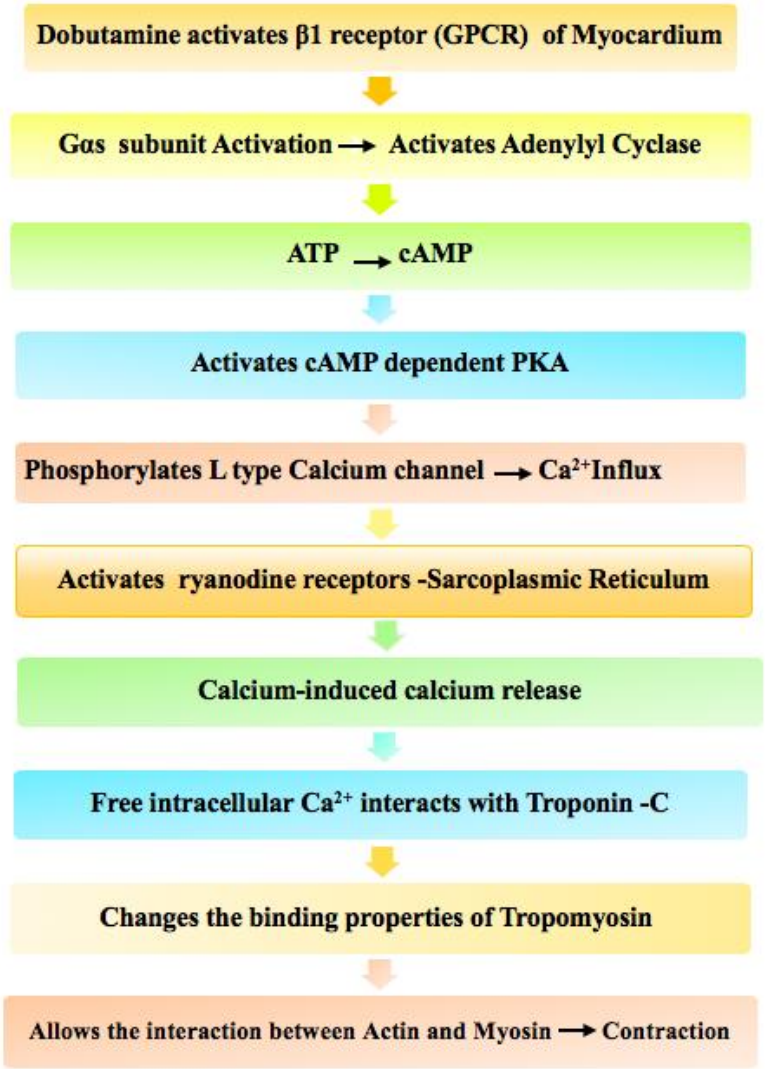


Fig 2 : Mechanism of Positive Inotropic Action of Dobutamine

Dobutamine - Pharmacological Actions :

Dobutamine by its strong β1receptor stimulant activity along with mild to moderate β2 receptor agonism and mild α 1receptor agonism produces a strong dose-dependent increase in Stroke Volume and Cardiac Output with modest increase in Heart Rate and a variable effect on Mean Arterial Pressure.Dobutamine causes moderate reduction in

Peripheral Vascular Resistance, except at high doses (>10-15 mcg/kg/min) when dose-dependent α1receptor agonism may become more prominent.The net effects of dobutamine on Mean Arterial Pressure depends on the relative changes in Cardiac Output and Peripheral Vascular Resistance from baseline values. However when Cardiac Output increases significantly and peripheral vascular resistance decreases moderately Dobutamine may raise Mean Arterial Pressure. On the contrary, Dobutamine may produce hypotension when CO increases moderately and Peripheral Vascular Resistance decreases significantly.[55]

Dobutamine causes a dose-dependent increase in HR, with low doses (up to 5 mcg/kg/min) producing increase in Stroke Volume via inotropic effects without significant tachycardia, but doses >10 mcg/kg/min causes worsening of tachycardia with negligible increase in Cardiac output any further due to falling Stroke Volume resulting in reduced diastolic filling time.[55]

Dobutamine – Pharmacokinetics :

Dobutaminehas a rapid onset of action (i.e) around 1 to 2 minutes after intravenous administration, with peak effectsobserved in 10 minutes. Due to its rapid metabolism, Dobutamine can be administered only by continuous intravenous infusion.Dobutamine has a half-life (t1/2) of 2 minutesand steady-state concentration is reached within 10 minutes that correlates with peak effect. Tachyphylaxis may occur with dobutamine infusions longer than 72 hours.[56,57]

Dobutamine undergoes methylation by Catechol-O-Methyltransferase (COMT). According to Raxworthy et al. , Dobutamine was found to be5-fold better substrate for catechol-O-methyltransferase than isoprenaline.[56]

Similarly Yan et al concluded that the main catabolic product of dobutamine is the 3-O-methyldobutamine. The isolation and identification of 3-O-methyldobutamine in the urine of individuals receiving infusion of racemic dobutamine was described by them. 47% of infused dobutamine was identified as 3-O-methyldobutamine while 33% was identified to be acid-hydrolysed derivatives that was mostly conjugated with sulphate. neonates.[57,58]

Yan et al.alsostated that the main catabolic fate of dobutamine in humans is the formation of 3-O-methyldobutamine. They described the isolation and identification of 3-O-methyldobutamine in the urine of children receiving infusion of racemic dobutamine. Forty-seven percent of infused dobutamine was identified as 3-O-methyldobutamine and its acid-hydrolysed derivatives, the latter mostly conjugated with sulphate (33%).Free plasma clearance of dobutamine is 102 ± 15 ml/kg/min. Renal excretion is mainly responsible for wide interindividual variability in plasma free dobutamine clearance rates.[56]

Leier et al. observed a linear relationship between the dose of dobutamine administered and the resulting plasma concentration. They also found a linear relationship to exist between the plasma concentration and the resulting hemodynamic response. The mean calculated threshold values signifying the minimum concentration needed for a change in cardiac outputwas 39 ng/ml.[56]

Dobutamine – Limitations:

An important limitation in the use of dobutamine is that in patients with heart failure, β adrenergic receptors may have undergone chronic downregulation hence may not produce full-fledged hemodynamic effects. Also, dobutamine causes increase in myocardial oxygen demand and oxygen consumption, that is deleterious in patients who have ischemic heart disease. Dobutamine at higher doses is also associated with higher rates of ventricular arrythmias. Tachyphylaxis to dobutamine effects may occur when dobutamine infusions are continued for more than 72 hourspossiblydue to induction of β-adrenergic receptor downregulation.[51,57]

Pulmonary Capillary Wedge Pressure (PCWP)

Pulmonary Capillary Wedge Pressure, is an amplitude-dampened form of Left Atrial pressure that is phase-delayed.[16,60]

It has two important implications.The first being that it provides an estimate of the hydrostatic pressure which is responsible for forcing fluid out of the pulmonary vascular space. The second implication is that the pulmonary capillary wedge pressure is directly proportional to the stretch of the fibre during diastole according to Starling's principle, which states that the force of contraction is proportional to myocardial fibre length/Left Ventricular volume. This serves to construct a cardiac function curve that is referred to as the LV filling pressure or preload.[16]

Measurement of PCWP : Swan-Ganz Pulmonary Artery catheter:

Introduced in 1972,the pulmonary artery catheter was named after its inventors Jeremy Swan and William Ganz.ThePulmonary Artery Catheter (PAC) helps to

monitor Pulmonary Artery Pressure(PAP), pulmonary capillary wedge pressure (PCWP) as well as Central Venous Pressure (CVP) easily.

The typical 7F thermo-dilution Pulmonary Artery Catheter (PAC)isa 110 cm long single catheter and consists of four lumina. It is made of flexible, radio-opaque polyvinyl chloride. Starting from the distal end of the catheter are marks in black at 10 cm increments. At the distal end of the catheter is a balloon made up of latex, capacity of which is 1.5 ml. On inflation, the balloon extends a little beyond the tip of the catheter however does not obstruct it. The distal balloon possesses the following three advantages (i) Prevents the catheter tip from making contact with the right ventricularwall during passage and hence reduces the incidence of arrhythmias during insertion (ii)Helps to float the catheter into the Pulmonary Artery (iii) Importantly inflation of the balloon allows the measurement of PCWP.[60]

At the tip of the distal end of the catheter, the lumen terminates. It is used to (i) measure chamber pressures; Pulmonary Artery Pressure& PCWP (ii) obtain samples of the mixed venous blood. Proximal catheter lumen ends 30 cm from the tip of the catheter, placing it in the right atrium (RA) while the distal opening is in the PA. Its advantages are that (i) it carries the injectate necessary for cardiac output computation (ii) can be used to measure the Central Venous Pressure (CVP) which indicates Right Atrial Pressure (iii) can be used to infuse vasoactive drugs. The third lumen has the electrical leads for thermistor, and is located at the catheter surfaceproximal to the tip by 4 cm.[61]

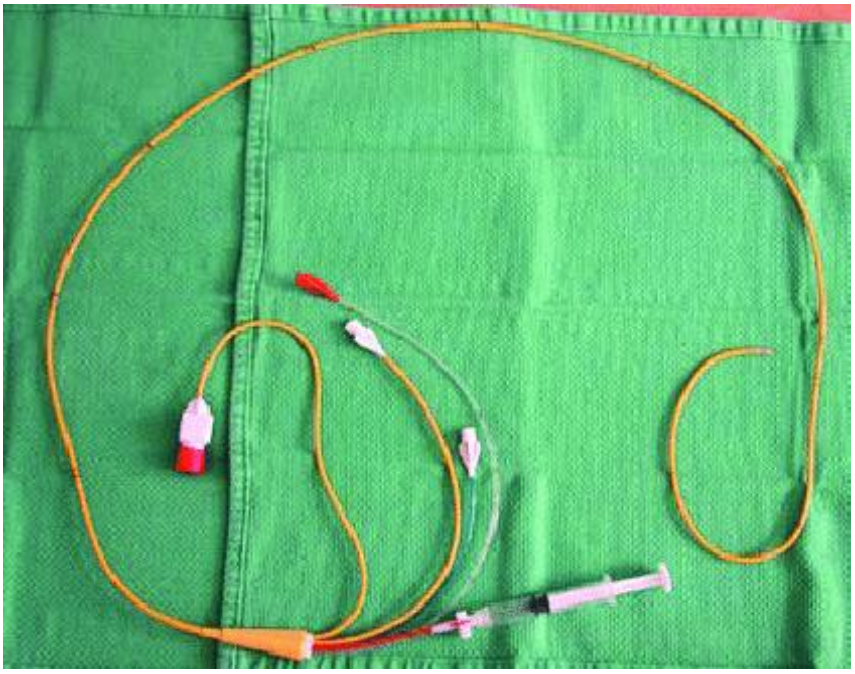


Figure 3 : Swan-Ganz Pulmonary Artery catheter

Insertion of Swan-Ganz Catheter:

The insertion of the Pulmonary Artery Catheter is an easy, fast and effective technique. For catheterizationa number of venous entry sites are employed.Anaesthesiologists and critical care physicianspreferthe Right Internal Jugular Vein (RIJV) approach for insertion of the PAC. From the RIJV catheter is advanced into the Superior Vena Cava and then into the Right atrium and the balloon is inflated with 1.5 ml of air and as the PAC is further advanced a dramatic change in the pressure tracing is producedwhen the tip of the catheter enters the Right Ventricle andpressure changes from that characteristic of RA into a phasic pressure in the range of 25 /0 mm Hg. From herethe catheter is introduced through the RV until it enters the main Pulmonary Artery which is marked byan increase in the diastolic pressure (25/12 mm Hg).On further advancement,the PAC wedges in a branch of the Pulmonary artery

.The PCWP is obtained when a catheter tip engages a small PA vessel (<2 mm).In the absence of any mitral valvular disorders PCWP reflectsthe left-atrial pressure with *a* , *v* waves and *x, y* descents that are transmitted from the Left atrium in a retrograde fashion(Figure 3). The PCWP ranges between 8-12 mm Hg .[17,60]

To obtain an accurate, high-quality PCWP tracing, an uninterrupted fluid column between the catheter tip and the left atrium is essential. The lung however consists of three distinct physiologic pressure zones, that have a different relation between the alveolar, pulmonary artery, and pulmonary venous pressures referred to as "The Lung Zones Of West". Zone 1 is characteristically present in the apex of the lungs, here the alveolar pressure is larger than the mean pulmonary artery and pulmonary venous pressures. Zone 2 is situated in the middle portion of the lung, here pulmonary artery pressure is greater than the alveolar pressure, which, in turn, is greater than the pulmonary venous pressure. Zones 1 and 2 are not suitable for estimation of the PCWP sincecollapse of capillary is present based on their pressure relations, and a direct column of blood does not exist between the left atrium and the wedged catheter tip. However, at the Lung zone 3 which is located at the base of the lung, the alveolar pressure is lower than both pulmonary arterial and pulmonary venous pressure, and is conducive for transmission of pressure directly from the left atrium to the wedged catheter tip. Therefore Lung zone 3 is the site where the PCWP reflects left-atrial pressureaccurately.[17]

PCWP measurement is especially vulnerable to errors in measurement. It is hence crucial to obtain the characteristic PCWP tracing.[63]Characteristics of a high-quality PCWP tracingare (1) Well-defined *a* and *v* waves (*a* wave is absent in atrial

fibrillation, and at low pressures phasic waves may not be distinct); (2) appropriate fluoroscopic confirmation with the tip of the catheter in the distal pulmonary artery and no apparent motion of the catheter with the balloon inflated; (3) an oxygen saturation obtained from the PCWP position more than 90%; and (4) observation of a distinct, abrupt rise in mean pressure when the balloon is deflated or the catheter is removed from the PCWP position to the pulmonary artery. Of all the above, obtaining an oxygen saturation more than 90% from the catheter tip is the most confirmatory sign of a true PCWP. Practically, the mean PCWP is around 0–5 mmHg lesser than the pulmonary artery diastolic pressure except when there is an increase in pulmonary vascular resistance.[62]

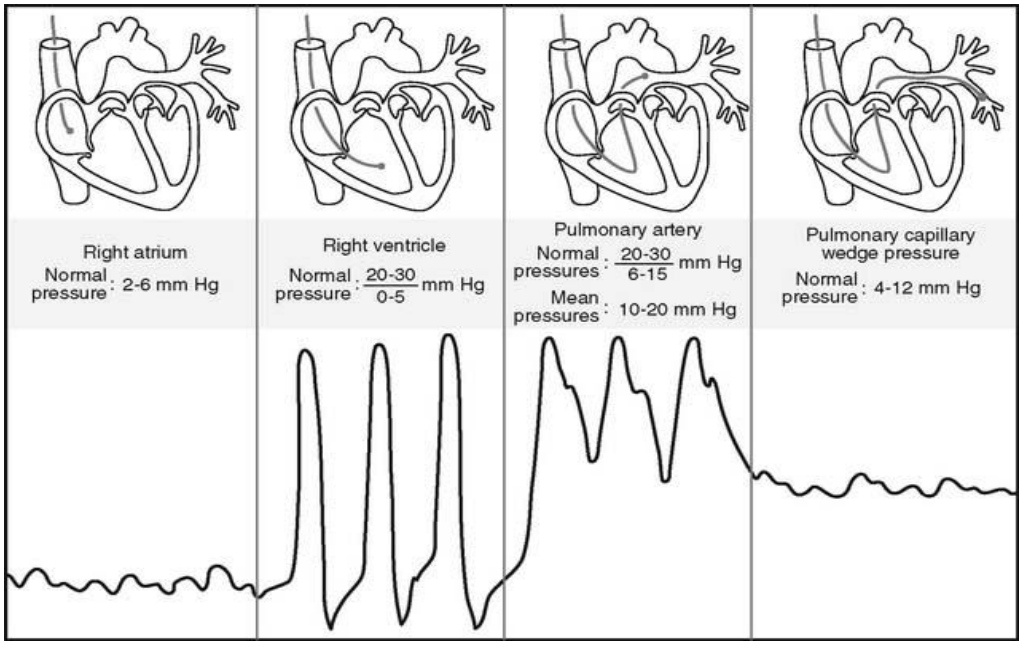


Figure 4: Sequence of pressures and pressure waveforms observed as the Swan Ganz Catheter advances through the right atrium, right ventricle, and pulmonary artery until it wedges.
Source: Heuer A, Scanlan CL. Wilkins' Clinical Assessment in Respiratory Care-E-Book. Elsevier Health Sciences; 2013 Aug 13.

PCWP and Low Cardiac Output Syndrome:

The LCOS that complicates CABG surgery is characterized primarily by LV failure leading to elevated LV end-diastolic pressure , which affects the left atrial pressure that in turn is transmitted into the pulmonary vasculature, causing pulmonary congestion.[66]The PCWP was found to correlate with the presence or absence of pulmonary congestion, whereas the CVP did not; an optimal cardiac output was associated with a PCWP of 15 mmHg.[67]

Pulmonary capillary wedge pressure (PCWP) is regardedas a surrogate estimate of left ventricular end diastolic pressure (LVEDP).PCWP represents an alternative measure to left ventricular end-diastolic pressure (LVEDP),which is the "gold standard" for determining Left Ventricular Filling Pressure (LVFP) .Pulmonary capillary wedge pressure plays a significant in the prevention and therapy of the low cardiac output syndrome.[64,65,68]

Need for Biomarkers:

Biomarkers are biological analytes which under specific physiological conditions can be detected in the blood . They serve as economically reasonable andreliable toolsin the screening, diagnosis as well as in prognosis of certain disease states.Despite controversies on the existence of a cardiovascular biomarker that can be deemed to be idealfor monitoring low cardiac output, an optimal biomarker would be highly sensitive and specific, released soon aiding early diagnosis, be persistent for a considerable amount of time that allows for a diagnostic window and can be

accurately as well as cost effectively quantified so that, it in turn would serve as a reflection of changes in patient's clinical status and prognosis. The biomarker could participate directly inthe pathogenesis or modulate disease. It could also be a by-product of the disease state. [69,70]

N (Amino)-Terminal -ProBrain(B-type) Natriuretic Peptide (NT-Pro BNP) :

Among biomarkers for cardiovascular disease states, Brain Natriuretic Peptide (BNP) and N-terminal pro-Brain natriuretic peptidehave evolved as powerful tools for diagnosis of acute heart failure as well as screening tools for identifying left ventricular systolic and diastolic dysfunction.[71]In a great number of studies both BNP and NT-Pro BNP have proven to be of diagnostic usefulness and have hence progressed from the bench to clinical application.[73]

A recent meta-analysis of 12 RCTs wasperformed by Savarese et al., on 2686 heart failure patients. It revealed that therapy guided by natriuretic peptide- is associated with outcome benefits.[72]

Source and Structure of NT-Pro BNP:

BNP was originally discovered in the porcine brain and was named porcine BNP, however further research revealed that the highest concentrations were found in the heart than in the brain.Human BNP is a polypeptide containing 32 amino acid that contains a 17 amino acid ring structure possessing a disulphide bond that connects two cysteine residues. BNP is predominantly synthesized and secreted by cardiac myocytes of the left ventricle when the myocytes undergo stretch as a result of

pressure overload or volume expansion of the ventricle. The gene encoding BNP in humans is present on chromosome 1, and the mRNA that encodes BNP has an unstable repeat TATTTAT sequence. The consequence of which is that rather than being stored in myocardial tissue under physiological states, the transcription of mRNA that encodes BNP leading to the synthesis and subsequent secretion of BNP occurs in an explosive manner. BNP is hence released immediately after synthesis in myocytes into surrounding tissues. In pathological states, the unstable mRNA can quickly synthesize a 134 amino acid BNP precursor (pre-proBNP) and cleaveaway the *N*-terminal 26 amino acid signal peptide to form a 108 amino acid BNP (proBNP), after which, proBNP is broken down by the proNP convertases, corin or furin, into a 76-amino acid biologically inert NT-proBNP and an active 32-amino acid BNP.The 108 amino acid precursor proBNP, biologically active BNP, NT-proBNPcanall be detected in plasma.[73]

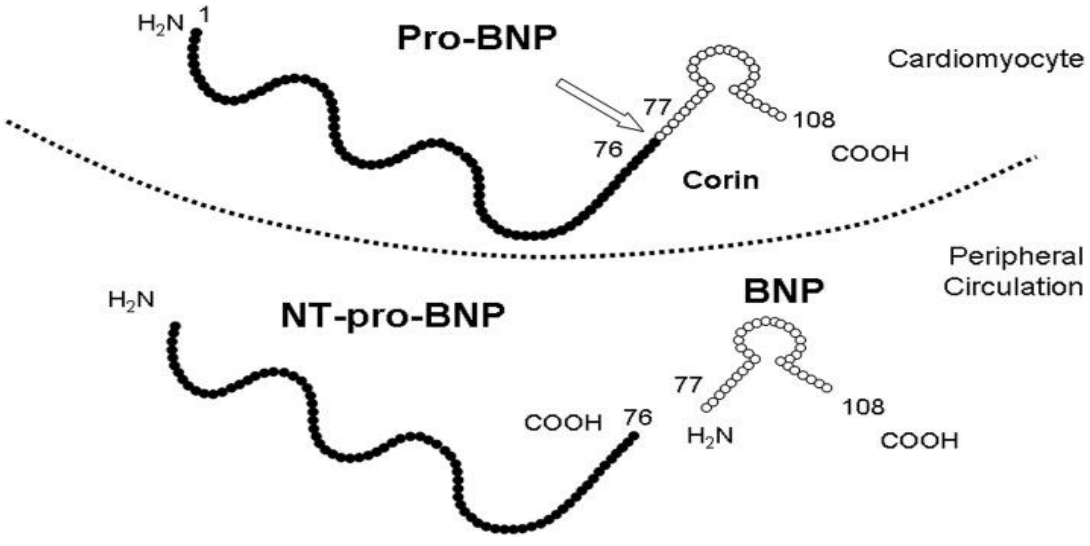


Figure 5: Molecular forms of Pro-BNP, NT-Pro BNP and BNP

Source:Troughton RW, Richards AM. B-type natriuretic peptides and echocardiographic measures of cardiac structure and function. JACC: Cardiovascular Imaging. 2009 Feb 1;2(2):216-25.

Degradation and elimination – Comparison of BNP with NT-Pro BNP :

Current research demonstrates that BNP is cleared from the circulation by a combination of action of Natriuretic Peptide Receptors -C (NPR-C), Neutral EndoPeptidase (NEP) mediated cleavage and inactivation and by normal glomerular filtration responsible for an estimated half-life of 20 min in comparison to 90–120 min for NT-pro-BNP which only undergoes renal glomerular filtration. In addition to the NEP, BNP is also cleared by means of proteolysis by other peptidases such as serine proteases, peptidyl arginine aldehyde proteases and kallikrein-like proteases. A complication in measuring BNP concentrations (or any biomarker that is degradable) is its sensitivity to degradation. Proteolytic cleavage of BNP may begin within circulation or immediately after blood collection making precise measurements of plasma levels difficult to achieve. However, NT-pro BNP due its virtue of being removed only by renal filtration remains for a longer duration in the circulation with a six fold higher plasma level in addition stability at room temperature is better when compared to BNP. [11]

Yeo et al., in their multicentric study found that recoverable levels of BNP falls immediately after phlebotomy, which could probably reflect the on-going activity of neutral endopeptidases in the blood sample or activation of the kallikrein system in the tube within which the blood sample is collected.On the other hand they found thatin comparison to BNP , NT-proBNP is remarkably stable after release, and the methods

used for its measurement are highly precise which is yet another major problem for several methods of BNP measurement. [74]

NT-Pro BNP As A Marker For Heart Failure:

The American College of Cardiology Foundation/American Heart Association (ACCF/AHA) and the European Society of Cardiology (ESC) guidelines state that,NT-proBNP is considered to be the most beneficial and precise biomarker for diagnosing Heart Failure and Cardiac dysfunction. In additionthey are also responsible for the determination of the severity, guiding relevant treatment strategies, and assessing the prognosis of heart disease.[75,76]

Heart failure is characterized by a decrease in stroke volume, resultingin inadequate cardiac output to meet the body's demands. In conjunction with this reduction in stroke volume there occurs an increase in filling pressure. This high filling pressure stretches the walls of the heart, causing BNP release.[11]

As a diagnostic biomarker, BNP can help arrive at a differential diagnosis. It helps to differentiate dyspnoea due to heart failure whereinNT-Pro BNP levels are high , from dyspnoea due to other causes wherein NT-Pro BNP levels are normal. Early detection of heart failure is crucial, especially in the elderly population, where misdiagnosis of heart failure can easily and rapidly lead to morbidity and mortality.[11,77,78]

Large number of studies have observed that NT-proBNP levels are consistently increased in patients with heart failure; further, the level of NT-proBNP has been found to be related to disease severity, as indicated by functional class (New York

Heart Association class), left ventricular systolic ejection fraction and left ventricular diastolic function.[79]

Independent of their diagnostic potential, a number of large-scale studies have clearly demonstrated that highNT-proBNP levels provide strong information on prognosis for an unfavourable outcome (eg., all-cause mortality, cardiovascular death, readmission to hospital or cardiac events) in patients with heart failure or asymptomatic left ventricular dysfunction.[80]As a marker for assessing severity or progress of heart failure, NT-Pro BNPis also used to monitor treatment response in acute HF patients, in whom levels would be expected to decrease. Currently, BNP is also recognised as an important indicator of hospital discharge and future prognosis. [77,78]

Dhaliwal et al.,studied regarding the most efficacious monitoring frequency and observed that multiple BNP measurements did not provide an advantage in terms of prognosis over a single measurement when considering ‘‘disease effect’’ or ‘‘disease modifiability.’’ [78]

Head-to-head comparison studies on the diagnostic performance of BNP and NT-proBNP in patients with heart failure and patients with asymptomatic left ventricular dysfunction reveal that both markers performed equally well, with almost identical areas under the receiver operating characteristics curves.[81]

Clinical Cut-offs ofNT-proBNP:

According to the ESC guidelines for the diagnosis and treatment of acute and chronic Heart failure in 2016, all patients with suspected acute HF must have their plasma natriuretic peptide levels (BNP and NT-proBNP) tested to help identify acute HF. In the non-acute setting , the upper limit of normal for NT-proBNP is 125 pg/mL, whereas in an acute setting, the cut-off value for NT-proBNP is 300 pg/mL[75].

The International Collaborative of NT-proBNP (ICON) study, stated that age-dependent cut-offs of NT-proBNP may be more useful for the diagnosis of HF. Acute HF can be excluded for general age-independent cut-off of 300 pg/mL. However, a diagnosis of HF should be made for patients who are less than 50 years of age with NT-proBNP levels > 450 pg/mL, patients who are between 50 and 75 years old with NT-proBNP levels > 900 pg/mL, and patients who are more than 75 years old with NT-proBNP levels > 1800 pg/mL [82].

Cardio Pulmonary Bypass (CPB) Machine /Pump – "Heart-Lung Machine" :

A milestone innovation in cardiac surgery was the development of the cardiopulmonary bypass machine (CPB). John Gibbon is considered to be the pioneer inthe use of heart lung machine and heralded the onset of modern day open heart surgery.[83]

The key function of the CPB machine is to maintain systemic circulation while the heart is being operated upon , when its chambers are open or there occurs severe cardiac dysfunction. The CPB also known as the heart-lung machine re-routes the

blood from the chambers of the native heart and the lungs and returns it to the arterial circulation. The CPB circuit consists of a reservoir, anoxygenator, blood pump,arterial filter , heat exchanger, a device that delivers cardioplegia and cannulaeconnected to one another by tubing of varied sizes.A blood pump completes the circuit which is responsible to generate flow. The venous blood from the pulmonary circulation is re-directed to the venous reservoirs by means of venous cannula. The blood pump also referred to as roller or centrifugal pump drives blood volume through a membrane oxygenator in the forward direction this enablesimmediate transfusion of oxygenated blood into the systemic circulation. The flow generated by the CPB needs to besufficient to ensure and sustain an adequate cardiac output, this is generally attained by maintaining a flow of 2.2 litres/minute/m^2 and a mean arterial pressure greater than 65 mm of Hg. This helps to prevent inadequate tissue perfusion characterised byincreasing metabolic acidosis, venous oxygen desaturation, EEG changes. Preservation of perfusion pressureat an optimal level is also essential so that organ structure and function are not compromised. The CPB may either be normothermic or hypothermic.[84]

Studies have demonstrated low rewarming after hypothermia to be associated withlesser neurocognitive dysfunction. Prior to separation from CPB, a proper team communication is vital. A checklist for various safety parameters such as optimal temperature, heart rhythm, acid-base status, electrolytes, ventilation, de-airing and patient position is very essential. Heparin is commonly used to maintain anticoagulation. Once the patient is stable off-CPB , the effect of heparin can

be reversed using protamine. Those patients who are unable to maintain an adequate cardiac output while weaning from CABG require inotropic support.[84,85]

Cardio Pulmonary Bypass and Systemic Inflammatory Response Syndrome (SIRS):

Systemic Inflammatory Response Syndrome (SIRS) is a condition wherein the process of inflammatory response is no longer localisedto the site of injury, ratherit disseminates throughout the entire circulation, damaging all vital organs and if severe in intensity and of significantly longer duration can lead to patient morbidity and mortality.[86]

Inflammation is a part of all stages of atherosclerotic development. It is both a cause and a consequence of ischemic heart disease. The employment of cardiopulmonary bypass (CPB) in cardiothoracic and vascular surgery leads to a well-known activation of the immunologic response. In some patients this immunological response that is triggered may be disproportionate to the stimuli causing SIRS.Thespectrum of severity in SIRS related to cardiac surgery, ranges from mild to an acute life-threatening syndrome characterised by acute multi-organ failure which has a mortality rate of 50–90%.[87]The primary mechanism that initiates SIRS while using CPB iscontact activation during CPB (Figure:6). Contact activation is a consequence that results when patient's blood is exposed to the artificial surfaces /materials within the CPB circuit. After the initial phase of protein deposition, coagulation factor XII (Hageman factor) undergoes activation. Activated factor XIIastimulates a number of cascade systems involving coagulation,fibrinolysis, kallikrein and complement activation.The pathway that is finally common to all these cascade systems results in

activation of platelets and especially neutrophils and monocytes causing dissemination of the inflammatoryresponse throughoutthe circulation.[83]

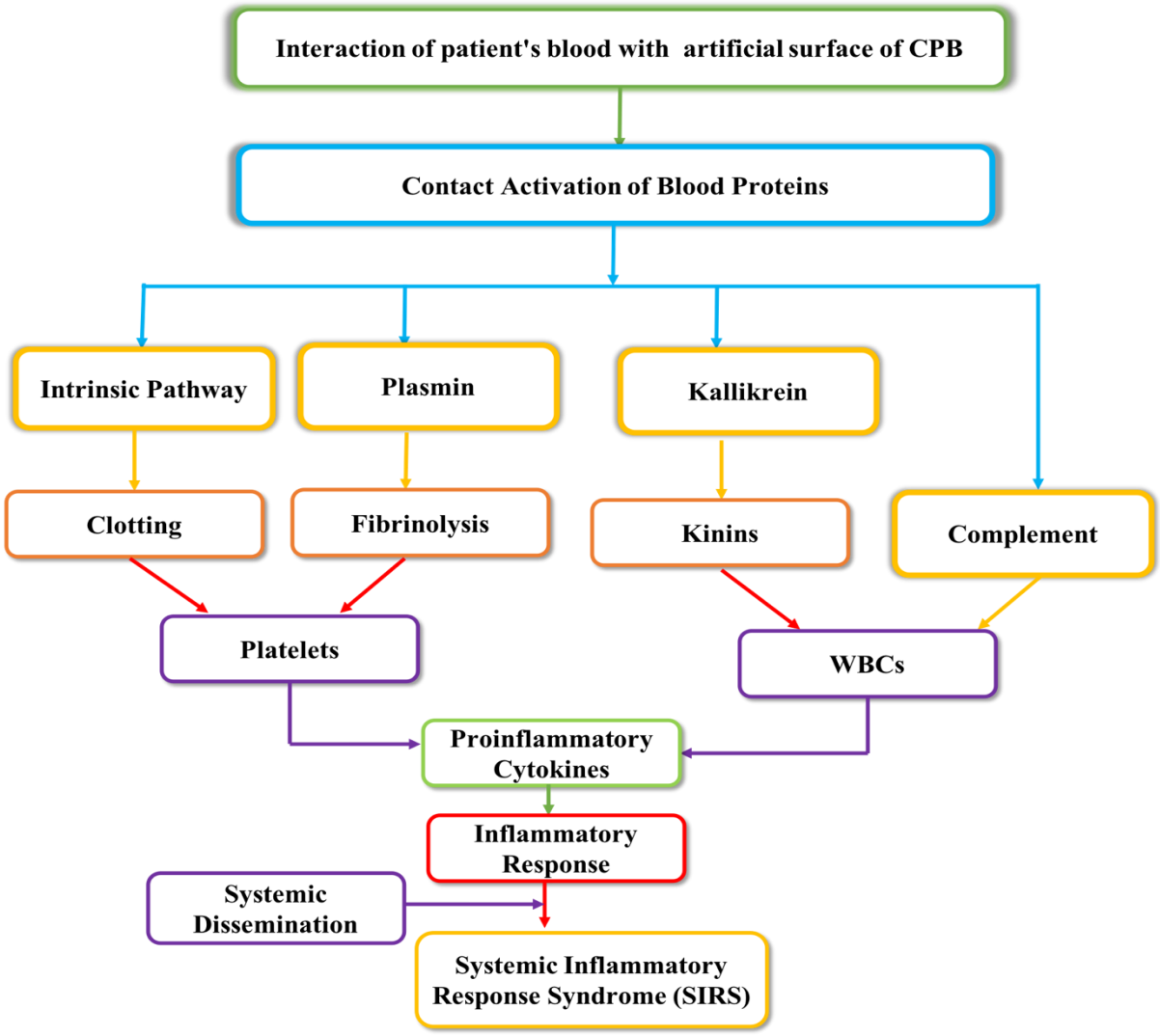


Figure 6 : Various pathways leading to inflammatory response following the use of CPB

Systemic Inflammatory ResponseSyndrome plays animportant role in perioperative complications.The release of different cytokines regarded as mediators of the process of inflammation, cellular activation, and leukocyte migration, is of prime importance.In very high concentrations, proinflammatory cytokines can affect the function of organs.On the other hand if anti-inflammatory effects predominate the

immune response would be blunted ,hindering defensive mechanisms and the process of healing.Hence a balance between pro and anti-inflammatory cytokines is essential.Of the pro-inflammatory cytokines, Tumour Necrosis Factor-α (TNF-α) and various Interleukins (IL)-1b, IL-6, IL-8are produced during the acute phase of the inflammatory response, and the high-sensitivity C-reactive protein (hs-CRP) that is released has been shown to correlate with multi-organ failure.[87-89]

Role of Tumour Necrosis Factor Alpha (TNF-α) in CABG surgery :

TumourNecrosis Factor-alpha is a proinflammatory cytokinethat plays a key role in the pathogenesis of the SIRS. TNF-α produced byinflammatory cells mediate various stages of inflammation and have the ability of stimulating many cells, including smooth muscle cells, fibroblasts, and endothelial cells.Tumor necrosis factor alpha acts by binding to specific receptors on cell surfaces. Various studies have demonstrated that TNF-α plays a major role in SIRS resulting from infection, burns, trauma, haemorrhagic shock, and pancreatitis. Tumor necrosis factor alpha participates in the outcome of other inflammatory processes, ischemia-reperfusion injury, delayed-type hypersensitivity, including allograft rejectionand granuloma development.When secreted in excess it may lead to organ dysfunction and death.[90]

In patients undergoing coronary revascularization surgery (CABG), in addition to CPB other stimuli such as general anaesthesia, surgical wounds, heparin administration, CPB, and protamine administration are also thought to play a role in the genesis of this response.[90]

Literature regarding the differences in inflammatory and cytokine responses between CABG surgery performed with and without CPB show that the levels of IL-6 were comparable in the two groups while Schulze et al., and Matasa et al., observed that levels of TNF-alpha were higher in patients undergoing on-pump CABG surgery in comparison to off- pump CABG surgery that is performed on a beating heart,hence confirming the role of CPB in elevation of TNF-alpha levels.[92,93]

Studies done previously on the increase of TNF-α levels in response to cardiac surgery at different time points confirmed that the levels reached a peak at 4 hours after surgery and degraded rapidly due to short half-life. [91]Matasa et al., also reported that the first step in the contact activation due to the use of CPB ,is the activation of complements and Polymorphonuclear cells, followed by the production of early proinflammatory (IL-8) cytokines and thereafter TNF. The elevated level of TNF may inturn cause decrease in contractile function and coronary flow that would further worsen the low cardiac ouptut syndrome complicating on-pump CABG. It was also observed that TNF-alpha levels normalized 6 hours after surgery .[93]

RATIONALE FOR THIS STUDY :

Although the correlation of NT-Pro BNP with PCWP has been established in both acute and chronic heart failure, to the best of our knowledge there are no studies till date evaluating the correlation of NT-Pro BNP values obtained preoperatively with PCWP measured intraoperatively in patients undergoing On-Pump CABG. If a correlation between the same exists, we consider it would help in risk stratification even prior to the surgery aiding us in early detection of those patients who will be prone to develop LCOS on table. Hence preoperative evaluation of NT-pro BNP levels in CABG patients could serve as an effective tool for predicting intraoperative complications necessitating haemodynamic support that could in turn affect the length of ICU stay and hospital stay. This could help physicians and surgeons in their decision making on determining if patients would be fit for surgery, if needed they could decide to delay on-pumpCABG surgery till the values are normalized.

CABG surgery using CPB machine results in a SIRS releasing many proinflammatory mediators among which TNF-alpha is found to be consistently elevated as per several studies and its association with reduced contractile function and postoperative morbidity and mortality have been established . However, there is a lack of studies correlating the intraoperatively measured plasma levels of TNF-alpha and plasma concentrations of dobutamine, which is the most commonly used inotrope during the course of On- Pump CABG surgery and in the perioperative period.

Hence we decided to do this study to evaluate the role of plasma NT-Pro BNP on PCWP during On-Pump CABG surgery and the role of TNF-alpha on the plasma concentration of dobutamine in the intraoperative period of patients undergoing On-pump CABG surgery to expand our knowledge on the role of these biomarkers in improving patient outcomes.

MATERIALS AND METHODS:

Study Design:

The study was designed as aprospective, clinical, observational, analytical study in patients undergoing on-pump CABG surgery.

Study population:

All patients undergoing elective On-Pump CABG surgery at Cardio Thoracic and Vascular Surgery Department of PSG Hospital, satisfying the eligibility criteria and provide written informed consent.

Study Duration :

One year time period between July 2018 and June 2019

Study Centre:

The study was performed at the Department of Pharmacology at PSG Institute of Medical Sciences & Research (PSG IMS&R) in association with Department of CTVS , PSG Hospitals, Coimbatore.

Sampling Method:

Consecutive Sampling

Sample size:

The sample size was decided to be 44.

Sample Size Justification :

As this is a first of its kind study, we were unable to obtain statistics from previous studies for sample size calculation and performed a convenience sampling. Between the time period of January 2017 and October 2017, 147 patients had undergone On-Pump CABG surgery in our CTVS department. The estimated annual number of CABG surgeries was calculated to be 176. A convenience sampling of 10% of the population that is usually done for such first of its studies yielded a meagre sample size of 17 patients . According to Hertzog M.A (2007) a sample size of 40 reduces error in estimation to 6 percentage points from 8 percentage points with 20 participants [94]. Hence we decided to perform a sampling of 25 % of the total number of estimated surgeries which yielded a **sample size *n* = 44**.

Study Approval :

The study protocol was approved by the Institutional Human Ethics Committee at PSGIMS &R(Proposal No. 17/370,dated 27.12.2017) before initiation of the study.

The study was also registered prospectively in Clinical Trial Registry of India – **CTRI No: CTRI/2018/05/013554 [Registered on 01/05/2018]**

Inclusion Criteria:

i. Patients undergoing elective On-Pump CABG surgery between July 2018 and June 2019
ii. Age : 40- 69 years
iii. Both sexes

iv. Co morbidities : Hypertension /Diabetes Mellitus /Thyroid dysfunction / Dyslipidaemia

v. Mild to Moderate Left Ventricular Dysfunction

Exclusion Criteria:

i. Patients undergoing Emergency CABG surgery

ii. Patients undergoing Off-Pump CABG Surgery

iii. Patients with preoperative unstable conditions such as: Severe Left Ventricular Dysfunction, Valvular Heart Disease, Pulmonary Arterial Hypertension, Abnormal Liver or Renal function

iv. Chronic anti-inflammatory therapy

v. Therapy with recombinant BNP analogues

Study Methodology (Figure:7) **:**

After obtaining approval for the study protocol from the IHEC and registering the study prospectively in CTRI, the study was commenced. Study participants who satisfied the inclusion and exclusion criteria were recruited and were thoroughly informed about the nature of the study. A written informed consent was obtained from all those individuals who were willing to participate in the study. Basic demographic details,anthropometric data , history related to duration of CAD, other comorbidities, drug intake, previous PCI/CABG history and substance use were collected.Baseline hemodynamic data, routinepre-operative lab investigations and baseline echo parameters that are done as a part of treatment protocol were also collected.

METHODOLOGY:

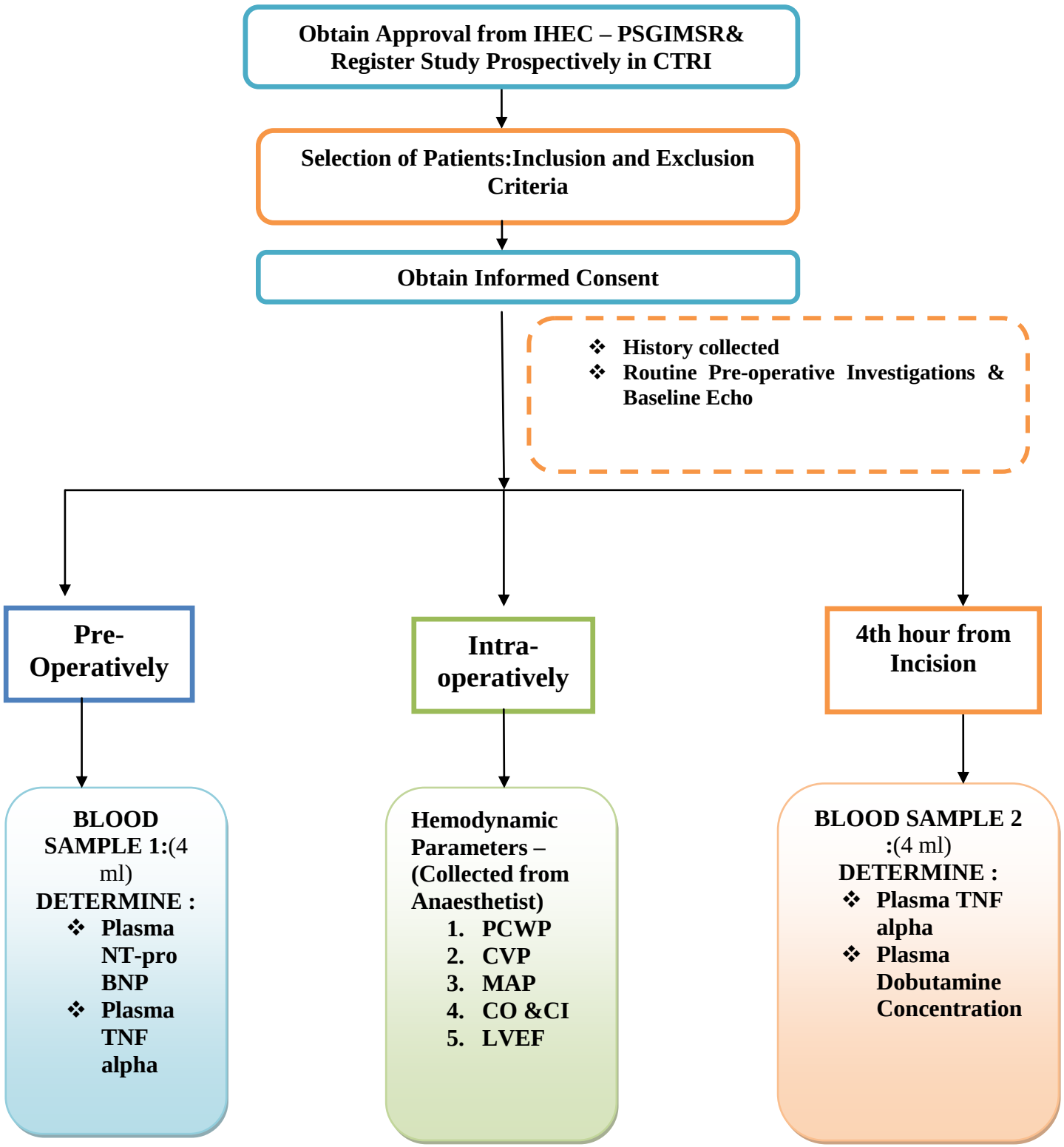


Figure 7 : Study methodology profile

Preoperative Sample Collection and Processing:

For each of the 44 patients , the day prior to on-pump CABG surgery 4 ml of venous blood sample was drawn and collected in sterile tubes made of polypropylene containing Ethylene Diamine Tetra Acetic acid (EDTA).This preoperative blood sample was centrifuged at 4°C for 10 minutes at 3000 rpm.The supernatant plasma was micro pipetted into two 2ml eppendorf tubes, one for quantitative analysis of preoperative NT-Pro BNP and the other for quantitative estimation of preoperative TNF-alpha concentrations in plasma. The eppendorf tubes with plasma were stored at -80°C till ELISA analysis.

On- Pump CABG Procedure:

Intraoperatively, anaesthesia was induced usingmidazolam 0.05 mg/kg, etomidate 0.1-0.2 mg/kg, vecuronium bromide 0.15 mg/kg, fentanyl 5-10 ug/kg. Following which central venous and Swan-Ganz catheters were placed. Anaesthesia was maintained using Propofol 6-10 mg/kg/h, atracurium 5-10 ug/kg/min, fentanyl intermittently.On-pump patients were managed using CPB equipment that comprised ofnon-pulsatileroller pumps (Stoeckert, Munich, Germany) and membrane oxygenators (Affinity,Avecor Cardiovascular, Plymouth, USA).The priming of the pump was done using a standardelectrolyte solution containing 5000 IUheparin, 1000 mL Ringer's lactate, 500 mLNaCl 0.9%, and 250 mL of a 15% mannitolsolution. Heparin(300 IU/kg) was administered immediatelybefore vascular cannulation. After the institution of CPB at a flow rate of 2.4–3 L/m^2 per min, the aorta was cross-clampedand a bloody

cardioplegic solutionwas injected.Once revascularization was completed, the effect of heparin was reversed using protamine sulphate (at a ratio of 1.5:1) in all patients.

Insertion ofSwan-Ganz Catheter to record PCWP:

The patient is positioned supine in 30°Trendelenberg position with head turned to the left side. The surgical area of the neck is prepared and draped. The Right Internal Jugular Vein is identified using a 22G needle. Following which a 18G thin walled 5 cm teflon catheter is placed into the vein and is threaded down the vessel for a short distance. The flexible end of the guide wire is passed through the 18 G catheter into the superior vena cava and the 18 G catheter removed. A dilator set is passed into the IJV over the guide wire by a twisting motion till the catheter sheath is in the SVC. Then the 7 F Swan Ganz catheter, that has been filled with fluid and attached to a transducer, is introduced through the sheath into the SVC. Correct positioning of the tip of the catheter in a central vein is confirmed by the changes in pressure in relation to respiration or coughing. When the catheter enters the RA, the balloon is inflated with 1.5 ml of air and the catheter is further advanced which in turn will produce a drastic change in the pressure tracing on entry into the RV .Changes in pressure from the characteristic RA to a phasic pressure in the range of 25 /0 mm Hg, typical of RV is identified. From the RV, the catheter is advanced further until it enters the main PA. This is identified by an increase indiastolic pressure (25/12 mm Hg) with no change in systolic pressure usually.Still further advancement of the catheter is advanced results in the catheter wedging into a branch of the Pulmonary Artery with a pressure pattern similar to atrial pressure pattern with a, c and v wave components transmitted

retrogradely from LA (PCWP=6-12 mm Hg) . The position of the PCW is verified by the characteristic waveform, a mean pressure lesser than the mean PAP and the ability to withdraw arterialised blood. Once the wedge position has been achieved, the balloon is deflated. This produces a typical PAP tracing.

Intraoperative recording of PCWP and other hemodynamic data:

Intraoperatively the anaesthetist records the PCWP,Mean Arterial Pressure,Systolic and Diastolic BP,Pulse rate,CVP,PAP,Cardiac output,Cardiac Index.These data are collected from the anaesthetist at the end of the procedure.In addition the anaesthetist's recordings of duration of CPB, duration of Aortic Cross Clamp, need for IABP(Intra AorticBalloon Pump) support are also noted.

Intraoperative Sample Collection and Processing:

For all the 44 patients undergoingon-pump CABG surgery, fourth hour from incision which according to literature review is the time for peak in TNF-alpha levels, venous blood sample of 4ml volume was drawn and collected in sterile tubes made of polypropylene containing Ethylene Diamine Tetra Acetic acid (EDTA).This intraoperative blood sample was centrifuged at 4°C for 10 minutes at 3000 rpm. The supernatant plasma was micro pipetted into two 2ml eppendorf tubes, one for quantitative analysis of intra operative TNF-alpha levels and the other for quantitative estimation of intraoperative steady state plasma concentration of the inotrope dobutamine. The eppendorf tubes with plasma were stored at -80°C till ELISA analysis for TNF-alpha and HPLC analysis for dobutamine levels.

Quantitative analysis of pre-operative and intra-operative TNF-Alpha levels using ELISA method :

Plasma TNF-alpha levels were measured usingsandwich Enzyme-Linked Immunosorbent Assay(ELISA), by a commercial kit (Bioassay Technology Laboratory,E0082Hu). It is based on the principle that the assay plate is pre-coated with human TNF Alpha antibody. When the sample is added, TNF Alpha present in it binds to the antibodies coated on the wells. Following which biotinylated human TNF Alpha Antibody is added that binds to TNF Alpha present in the sample. Then Streptavidin-HRP is added that in turn binds to the Biotinylated TNF A antibody. After incubation, the unbound Streptavidin-HRP is washed away in the washing step. When substrate solution is added a colour develops that is in proportion to the amount of human TNF Alpha in the sample. The addition of acidic stop solution terminates the reaction and the absorbance is measured at 450 nm.

- The reagents used for the analysis are as follows :

1. Standard Solution (960ng/L)
2. Pre-coated ELISA Plate
3. Standard Diluent
4. Streptavidin-HRP
5. Stop Solution
6. Substrate Solution A
7. Substrate Solution B
8. Wash Buffer Concentrate (30x)

9. Biotinylated human TNF A Antibody
10. User Instruction
11. Plate Sealer
12. Zipper bag

- Reagent Preparation :
  - All reagents were brought to room temperature before use.

  - **Standard :**

    Of the standard(960ng/L) that was provided 120µl wasreconstituted with 120µl of the standard diluent to generate a 480ng/L of standard stock solution. The standard is allowed to sit for 15 mins with gentle agitation prior to making dilutions. Duplicate standard points were prepared by serially diluting the standard stock solution (480ng/L) 1:2 with standard diluent to produce 240ng/l, 120ng/L, 60ng/L and 30ng/L solutions. Standard diluent is considered as the zero standard(0 ng/L).

Concentration	Standard	Dilution
960ng/L	Original Standard	-
480ng/L	Standard No:5	120µl Original Standard + 120µl Standard Diluent
240ng/L	Standard No:4	120µl Standard No.5 + 120µl Standard Diluent
120ng/L	Standard No:3	120µl Standard No.4 + 120µl Standard Diluent
60ng/L	Standard No:2	120µl Standard No.3 + 120µl Standard Diluent
30ng/L	Standard No:1	120µl Standard No.2 + 120µl Standard Diluent

Table 1: Dilution of standard solutions

ELISA Assay Procedure:

1. All reagents, standard solutions and samples were brought to room temperature before use. The assay was performed at room temperature.
2. 50µl of standard was added to the standard well.
3. 40µl of sample was added to sample wells and then 10µl of anti-TNF A antibody, 50µl streptavidin-HRP were added to sample wells and standard wells but not blank control well and was mixed well. The platewas covered with a sealer andincubatedfor 60 minutes at 37°C.
4. Sealer was removed and the plate was washed 5 times with wash buffer. Wells were soaked with at least 0.35 ml wash buffer for 30 seconds to 1 minute for each wash. The plate was blotted onto paper towels

5. 50µl of substrate solution A was added to each well and then 50µl of substrate solution B was added to each well. The plate was covered with a new sealer and incubated for 10 minutes at 37°C in the dark.
6. 50µl of Stop Solution was added to each well, after which the blue colorchanged to yellow immediately.
7. The optical density (OD value) was determined for each well using a microplate reader (BIO-RAD)immediately that was set to 450 nm within 10 minuets of adding the stop solution.

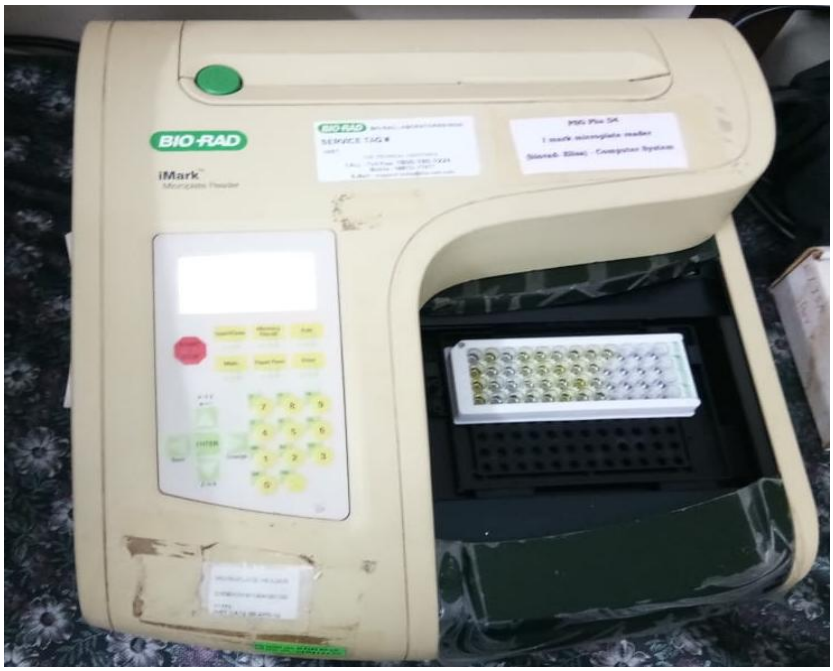


Figure 8 : Quantification of TNF Alpha using ELISA microplate reader

Quantitative analysis of pre-operative levels NT-Pro BNP using ELISA method :

Plasma NT-Pro BNP levels were measured by a commercial kit (Fine-Test, Wuhan Fine Biotech Co., Ltd., EH0350) using sandwich Enzyme-Linked Immunosorbent Assay. It was based on the principle of sandwich ELISA technology. Anti- NT-

proBNP antibody were pre-coated onto 96-well plates. Following which biotin conjugated anti- NT- ProBNP antibody was used as detection antibodies. The standards, test samples and biotin conjugated detection antibody were added to the wells sequentially, and were washed with wash buffer. Next , HRP-Streptavidin was added and unbound conjugates were washed away with wash buffer. TMB substrates were used to visualize HRP enzymatic reaction. HRP catalysed TMB to produce a blue color product which changed into yellow after adding acidic stop solution. The density of yellow is proportional to the NT-ProBNP concentration of sample captured in plate. The O.D. absorbance at 450nm was read in a microplate reader, and then the concentration of NT- ProBNP was calculated.

- **Reagent Preparation** :

30mL of concentrated wash buffer was diluted into 750 mL wash buffer with deionized or distilled water.

- **Standard:**
- 2500pg/ml of standard solution: 1 ml Sample / Standard dilution buffer is added into one Standard tube.
- Dilution from 1250pg/ml→39.063pg/ml of standard solutions:

6 Eppendorf tubes were labelled with 1250pg/ml, 625pg/ml, 312.5pg/ml, 156.25pg/ml, 78.125pg/ml, 39.063pg/ml, respectively. 0.3 ml of the Sample/Standard dilution buffer was added into each tube. 0.3 ml of the above

2500pg/ml standard solution was added into the 1st tube and was mixed thoroughly. Next from 1st tube0.3 ml was added to 2nd tube and so on.

Assay Procedure

1. 0.1ml of 2500pg/ml, 1250pg/ml, 625pg/ml, 312.5pg/ml, 156.25pg/ml, 78.125pg/ml, 39.063pg/ml, standard solutions were added into the standard wells.
2. 0.1 ml of Sample/standard dilution buffer was added into the control (zero) well.
3. 0.1 ml of plasma was added into test sample wells.
4. The plate was sealed with a cover and incubated at 37 °C for 90 minutes.
5. The plate was washed 2 times with Wash Buffer.
6. 0.1 ml of Biotin-labelled antibody working solution was added into above wells .
7. The plate was sealed with a cover and incubated at 37°C for 60 min.
8. The cover was removed and the plate was washed 3 times with Wash Buffer.
9. 0.1 ml of SABC Working Solution was added into each welland incubated at 37°C for 30 minutes.
10. Cover removed and plate washed 5 times with Wash Buffer.
11. 90µl TMB Substrate was added into each well and incubated for 15-30 mins at 37°C in dark. Following whichthe first 3-4 wellswith most concentrated NT-ProBNP standard solutions turned blue, while the other wells may not display obvious color.

12. 50µl Stop Solution was added into each well and mixed thoroughly. The color changes to yellow immediately.
13. The O.D. absorbance was read at 450 nm in Microplate Reader immediately after adding the stop solution.

Dobutamine Quantification : HPLC Analysis

Instrumentation and Chromatography Conditions

The High Performance Liquid Chromatography (HPLC) instrument consisted of a Waters acquity-H class HPLC system with a quaternary pump and 96-vial autosampler that is coupled with diode array UV detector (Waters,Millford,MA,USA). Separation by chromatography was performed onSun Fire column 120, EC C18 column from Waters (150mm x 4.6mm ; 2.7µm).The temperature of the column was set at 40°C.

Mobile Phase Preparation :

The mobile phase consisted of a mixture of methanol and water (20:80) v/v with 0.9ml of triethylamine at a pH of 3.0 adjusted using orthophosporic acid at a flow rate of 0.5ml/min. Prior to analysis the mobile phase was passed through 0.45µm membrane filter and degassed with ultra-sonication. A 20µl of each sample was injected in to the system and total run time was 5min with wavelength of E_x200 E_m330nm. Data acquisition wasperformed by Empower 2 software (Waters).

Preparation of standard stock solution of Dobutamine (1mg/ml):

10mg of API standardized dobutamine was weighed accurately and transferred into a dried 10ml volumetric flask. It was diluted with small amount of methanol and made up using water followed by sonication for 10 minutes to obtain the working standard solutions of dobutamine.

Preparation Internal Standard (IS) stock solution Dopamine (1mg/ml):

10mg of API standardized dobutamine was weighed accurately and transferred into a dried 10ml volumetric flask. It was diluted with small amount of mobile phase and made up to volume followed by sonication for 10minutes to get the working standard solutions of Dopamine.

Sample preparation:

Sample preparationwas carried out using liquid-liquid extraction method. 250µl of human plasmasample that was spiked was taken in 2ml Eppendorf tube. 10µL of IS Working standard solutions were mixed for30 seconds on a vortex to which 1.3ml of methyl tert-butyl ether was added.The contents of the tube were mixed on a vortex mixer for 3minutes and the tubes were centrifuged at 10000 rpm for 15minutes. 1ml of the organic layer was pipetted out into separate tube and evaporated to dryness at 60˚C using nitrogen evaporator . The residue was reconstituted using mobile phaseand was subjected to chromatographic analysis using the optimized chromatographic conditions.

Column	Sun fire 120, EC C18 (150mm x 4.6mm ; 2.7µm)
Flow rate	0.5ml/min
Wavelength	E_x200 E_m330
Temperature	40°C
Injection volume	20µl
Mobile phase	Methanol: phosphate buffer (20:80) PH (3.0)
Run time	6 min
Mode operation	Isocratic

Table 2: Optimized Chromatographic Conditions

Statistical Analysis:

Data of all the 44 patients who were enrolled in the study were included in the statistical analysis. All theirdemographical and clinical data were analysed usingIBM SPSS Statistics Software (Version 24.0 SPSS Inc., Chicago, IL, USA).

All categorical data were represented by frequency distributionswhile descriptive statistics were used for numerical data. To determine the normality of the numerical data, Shapiro-Wilk test was used.Box-whisker plot were used to represent the normality of data graphically.

Based on the normality assumptions, either parametric or non-parametric statistical tests of significance were performed in the data analysis. When data was found to follow normal distribution, the presence of a statistically significant difference between the two groups was determined using Independent sample t test. Analysis of variance (ANOVA) is used to find the presence or absence of statistically significant difference between more than two group data when the data follows normal distribution. For independent sample t test and ANOVA level of statistical significance was set at 5 % i.e $p < 0.05$

Pearson correlation analysis was used to analyse the correlation between NT-Pro BNP levels and PCWP as well as the correlation between TNF-alpha levels and Dobutamine concentration for which thestatistical significance was set p value<0.05fora confidence interval of 95%. The linearity is represented using a scatter plot.

RESULTS:

Study Participants:

Totally, 44 patientsaged between 40 and 70 years undergoing on-pump CABG surgery at PSG hospitals, fulfilling eligibility criteria were included in the study after obtaining written informed consent.

Baseline Demographic Profile:

The mean age of the study population was 57.02±7.70 years .The mean Body Mass Index (BMI) and Body Surface Area (BSA) were26.80±4.53 and1.57±0.14 m^2 respectively. The mean duration of hypertension was8.32±3.30 years ,while the mean duration of Diabetes Mellitus was7.47±2.99 years .The study population had a mean duration of Coronary Artery Disease (CAD), dyslipidaemia and thyroid dysfunction of9.51±2.32 years, 8.19±2.75 years and 5.00±2.00 years respectively (Table 3).The majority of study participants were males (64%) in comparison to females (36%) (Figure 9). The age wise distribution shows thatmost of the participants belonged to 61-70 years age group (46%) followed by participants in the 51-60 years age group (29%)and 41-50 years age group (25%) (Figure 10). Majority of participants hailed from urban areas (68%) and the restfrom rural residence(Figure 11).

Table 3 : Baseline demographic profile

	Mean ± SD / n(%)
Age (years)	57.02±7.70
Body Mass Index (BMI)	26.80±4.53
Body Surface Area (m^2)	1.57±0.14
Hypertension Duration (years)	8.32±3.30
Diabetes Mellitus Duration (years)	7.47±2.99
Thyroid Duration (years)	5.00±2.00
Dyslipidemia Duration (years)	8.19±2.75
CAD Duration (years)	9.51±2.32

Figure 9: Sex distribution in study population

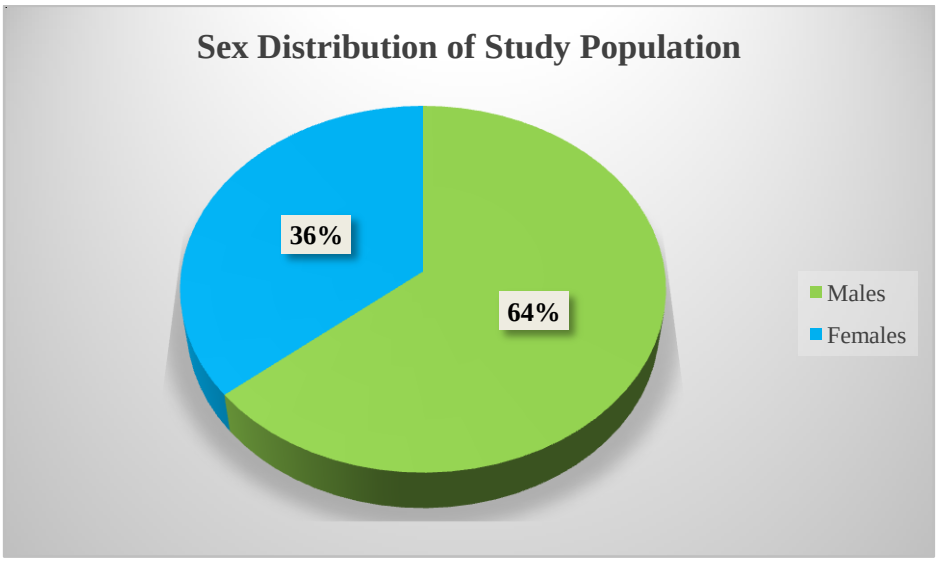


Figure 10: Age group wise Distribution of Study Population

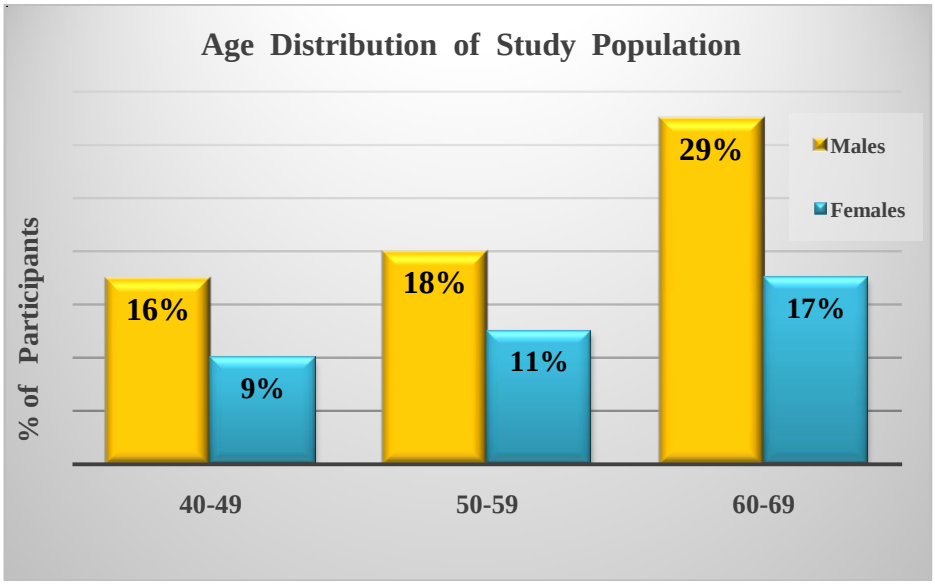
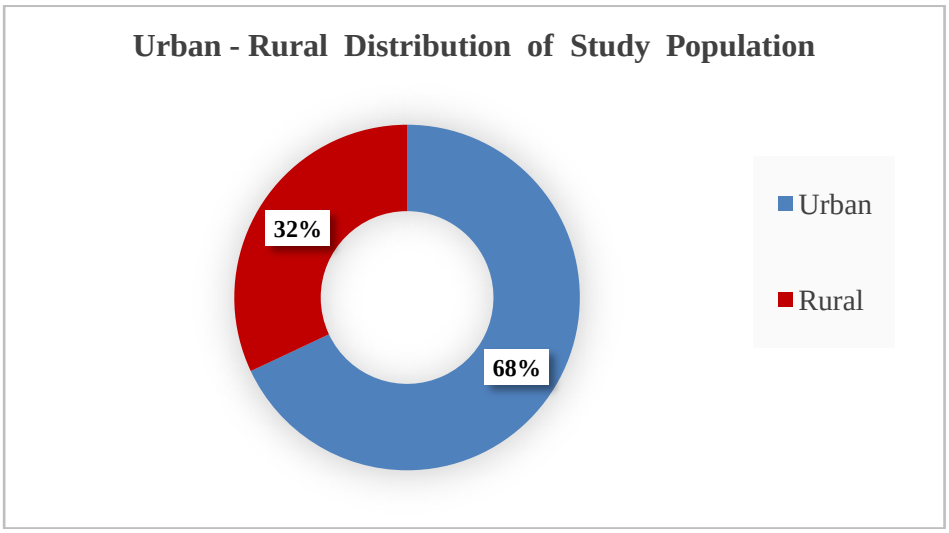


Figure 11:Urban – Rural Geographic Distribution of Study Population



It was observed from the baseline BMI distribution of the study participants that, the majority of the participants were pre-obese patients (34%) followed by obese patients (25%) and normal weight patients (25%). 16% of the participants were overweight. (Figure 12)

Figure 12:Baseline BMI Distribution of Study Population

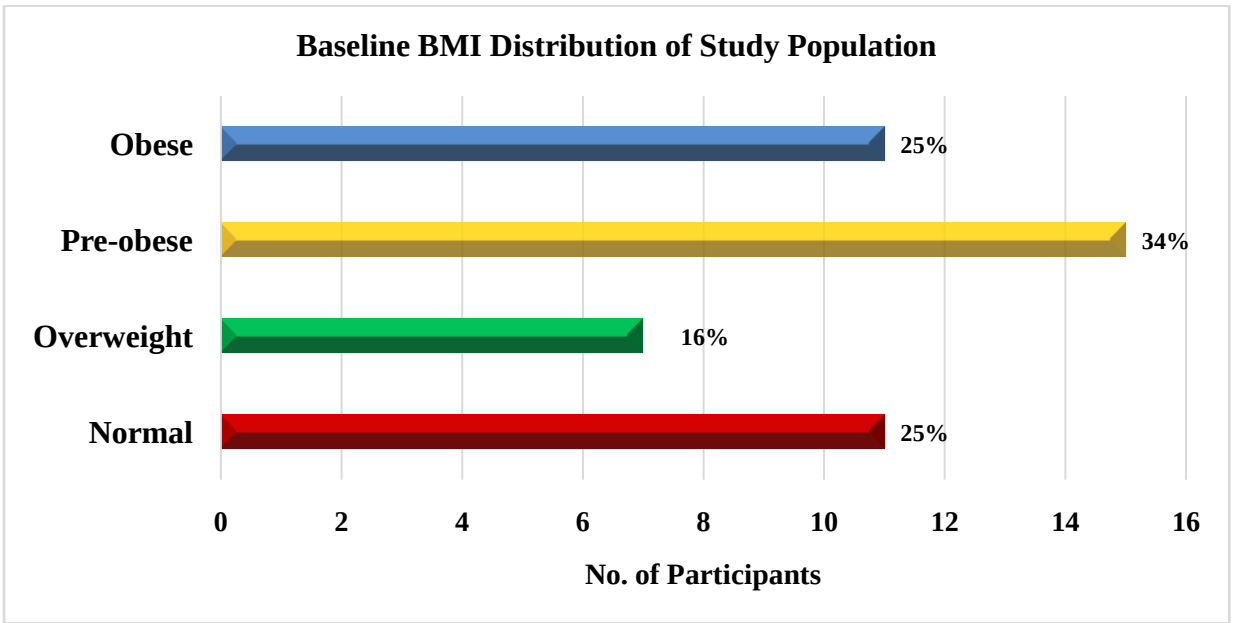


Table 4: Comparison ofAnthropometric Measurement And History Of Diseases Among Male And Female Patients

		Male Mean±SD (or) n (%)	Female Mean±SD (or) n (%)	P-value
BMI(kg/m2)		26.30±4.03	27.69±5.32	0.335
Body Surface Area (m2)		1.58±0.13	1.56±0.17	0.627
Diabetes	Present	24 (55%)	9 (20%)	0.067
	Absent	4 (9%)	7 (16%)	
Hypertension	Present	23 (52%)	13 (30%)	1.000
	Absent	5 (10%)	3 (7%)	
Thyroid	Hypothyroid	6 (14%)	3(7%)	1.000
	Absent	22(50%)	13(30%)	
Dyslipidemia	Present	24(55%)	15(34%)	0.638
	Absent	4(9%)	1(2%)	
Pre-existing Left Ventricular Dysfunction	Mild	16(36%)	11(25%)	0.447
	Moderate	12(27%)	5(11%)	
STEMI at presentation	Yes	3(7%)	1(2%)	1.000
	No	25(57%)	15(34%)	
History of previous MI	Yes	4(9%)	1(2%)	0.638
	No	24(55%)	15(34%)	
History of previous Stroke	Yes	4(9%)	0	0.280
	No	24(55%)	16(36%)	
History of peripheral vascular disease	Yes	4(9%)	2(4%)	1.000
	No	24(55%)	14(32%)	

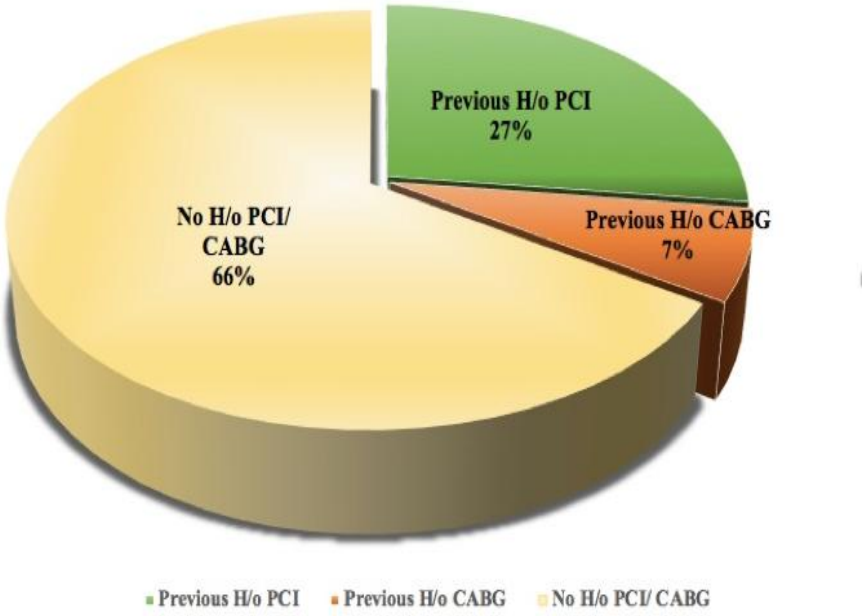
Comparison of anthropometric measurements and previous disease historybetween male and female participantsat 5% level of significance revealed that there was no statistically significant difference in terms of BMI and BSA between male and female participants with the mean in both groups falling under the pre-obese category.In addition, there was no statistically significant difference in the proportion of co-morbidities present among male and female participants. Majority of the male and female patients had a positive pasthistory of diabetes mellitus, hypertension, hypothyroid, dyslipidemia and left ventricular dysfunction. Whereas most of the male and female participants had a negative past history of MI, stroke and peripheral vascular disease. None of the female participants had a previous history of stroke but this was not statistically different from the proportion observed among males. (Table 4)

From the table 5 it was observed that, majority of the patients (98%) used Aspirin followed byAngiotensin Converting Enzyme Inhibitor /Angiotensin Receptor Blocker (ACEI/ARB) (96%) and Nitrates (90%) . Use of statins was observed among 66% while 57% of the study population used Calcium Channel Blockers (CCB).Among the drugs used for the treatment of diabetes mellitus it was observed that 57% of the participants were on oral antidiabetic drugs while 48% were on insulin. The use of diuretics was observed among 41% and the use of aldosterone antagonist among 39% of the participants.

Table5: Baseline Drug History among Study Population

Drugs	No. of patients	% of patients
Aspirin	42	98
Nitrates	40	90
Statins	38	86
CCB	29	66
Beta blocker	21	48
ACEI/ARB	43	96
Diuretics	18	41
Aldosterone antagonist	17	39
Oral Antidiabetic drugs	25	57
Insulin	21	48

Fig 13: Baseline Frequency Distribution of Past History of Coronary Revascularization



A major proportion of participants (66%) did not have any prior history of PCI / CABG.Among those who had a previous history of Coronary Revascularization, most of them hadundergone PCI (27%) in the past whereas only a minor proportion (7%) had a previous history of CABG, now requiring a re-surgery in the present.(Figure 13)

It was observed that the use of alcohol (89%) was more common among the study population followed by the use of tobacco(64%). (Figure 14)

Figure 14:Baseline Distribution Of Substance Use In The Study Population

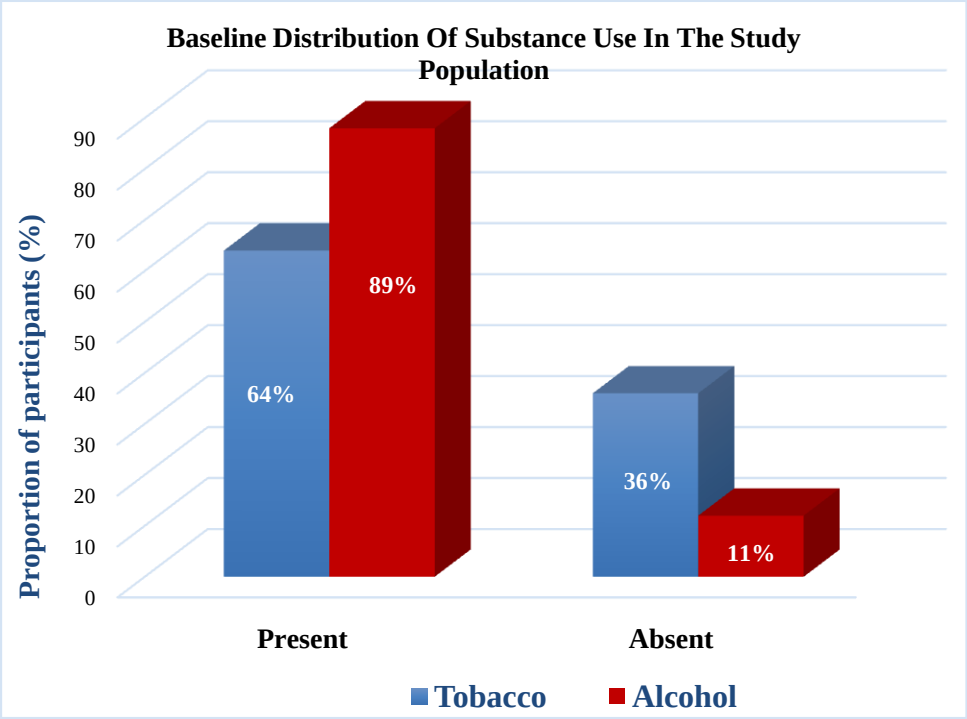


Table 6: Baseline laboratory values of the study population

Lab Measurements	Minimum	Maximum	Mean ± SD
Hb(g%)	11.20	17.90	14.42±1.72
HbA1C(%)	5.20	8.40	6.34±1.26
Serum urea(mg/dl)	14.90	38.20	25.78±5.72
Serum creatinine(mg/dl)	.40	1.20	0.79±0.23
Total Cholesterol(mg/dl)	152	312	215.77±37.59
Serum TGL(mg/dl)	124	256	187.27±33.44
CK-MB (ng/ml)	2.12	7.50	4.14±1.42
Troponin-Ths (pg/ml)	12.00	1200.14	78.39±202.78
Plasma NT-Pro BNP (pg/ml)	202.22	1056.00	685.37±255.64
Plasma TNF-Alpha(pg/ml)	5.70	35.60	17.9209±7.04

At baseline it was observed that the mean values of Hemoglobin, HbA1C, Serum Creatinine, Total Cholesterol, CK-MB and Plasma TNF-alpha in the study population were within normal limits. However, with regard to the mean levels of baseline serum urea, serum Triglycerides (TGL) and NT-Pro BNP it was seen that these lab parameters were abnormally elevated. The baseline mean Troponin-T hs levels revealed that on an average participants were in the category of intermediate risk of Acute Myocardial Infarction (AMI).(Table 6)

Table 7: Baseline Echocardiographic parameters of the study population

Echocardiographic parameters	**Mean ± SD / n(%)**
Left Ventricular Ejection Fraction (LVEF)	56.34±7.42
Regional Wall Motion Abnormality(RWMA)	22 (50%)
Left Ventricular Systolic Dysfunction (LVSD) Mild n(%)	26 (59.1%)
Left Ventricular Systolic Dysfunction(LVSD) Moderate n(%)	10 (22.7%)

On an average, the study participants had a normal LVEF. However, Left Ventricular Systolic Dysfunction(LVSD) was observed in 59% of patients at a mild Level and 23% of participants at a moderate level. In addition half of the study population had Regional Wall Motion Abnormality(RWMA) in their echocardiographic evaluation.(Table 7)

HPLC Analysis of Dobutamine Concentration in Plasma

Using High Performance Liquid Chromatography (HPLC), the plasma concentration of Dobutamine was analysed and a calibration curve was constructed. This calibration curve demonstrated excellent linearity with regression correlation coefficient (r^2>0.99) with a Limit of Detection (LoD) of 0.74 µg/ml and Limit of Quantification of(LoQ) of 1.18 µg/ml.LLoQ was the lowest concentration with RSD <20%. The concentration range was that exhibited linearity was 5- 1000 ng/ml.Across the calibration range, the standard calibration curve had a consistent reproducibility for the standard concentrations. The best fit of peak area ratio (Peak area analyte /Peak area IS) vs Concentration was determined and a typical regression equation was formulated and it was fitted to y = mx+c using a weighing factor ($1/x^2$).The percentage accuracy 99.26±0.29 % .(Figure 15-17)

The mean plasma concentration of dobutamine at 4th hour from incision was found to be 3.64µg/ml. (Table 8)

Figure 15:Representative Chromatogram of internal standard in patient plasma (1000.0 ng/mL)

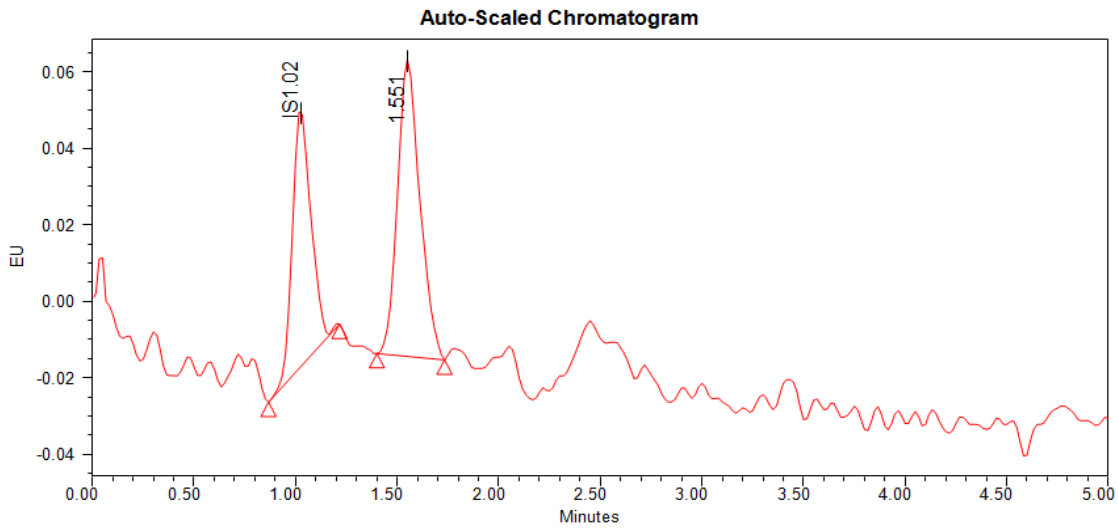


Figure 16: Representative Chromatogram of Dobutamine in patient Sample

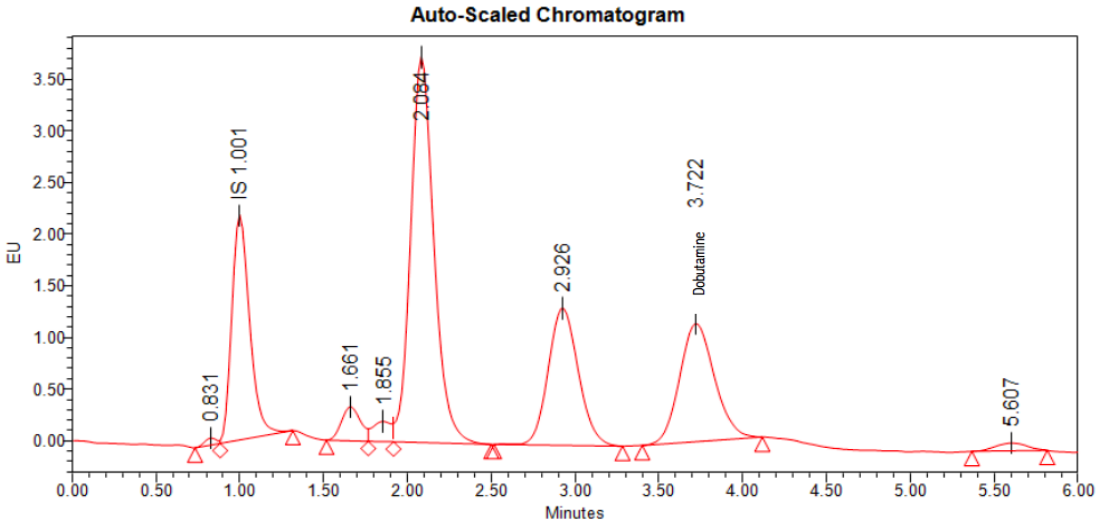


Figure 17: Representative Calibration Curve of Dobutamine in Plasma

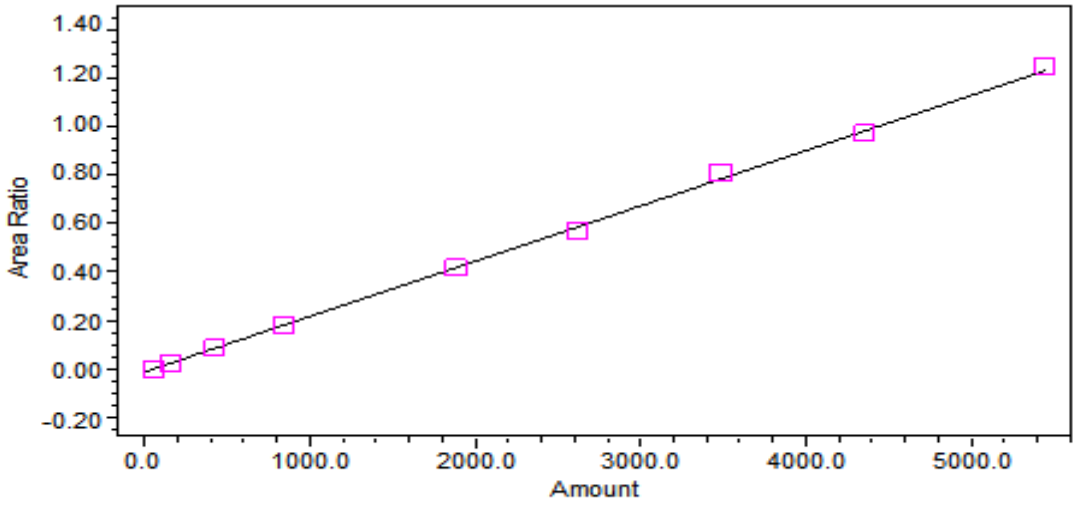


Table 8: Dobutamine Concentration and Absorbance area at 4th hour from incision

	Mean ± SD
Plasma Concentration of Dobutamine (μg/ml)	3.640 ± 0.42
Absorbance Area (mAu/min)	64399.31 ± 7389.23

Table 9: Intraoperative profile of the study population undergoing On-Pump CABG Surgery

	Mean± SD / n(%)
Hemodynamic Details	
Heart Rate (beats per minute)	76.95 ± 10.90
Systolic BP (mm Hg)	114± 11.81
Diastolic BP (mm Hg)	73 ± 9.71
PCWP(mm Hg)	12.27 ± 4.43
CVP(mm Hg)	5.68 ± 1.47
$ETCO_2$(mm Hg)	33.75 ± 2.42
Intra-operative Details	
CPB Time (minutes)	98.23 ± 9.91
Aortic Cross Clamp (ACC) Time (minutes)	60.20 ± 4.31
Antegrade Cardioplegia	44 (100)
Need for IABP Support	6 (14)
No.of Grafts	3 ± 0.81

Intraoperative details of the patients undergoing CABG surgery revealed that among the hemodynamic parameters, mean heart rate was 76.95 ± 10.90 bpm, while the mean systolic and diastolic BP were 114 ± 11.81 and 73 ± 9.71 mm of Hg respectively. The mean PCWP was 12.27 ± 4.43 mm of Hg.It was observed that all the hemodynamic

parameters were within normal limits.The mean $ETCO_2$ was33.75 ± 2.42 mm of Hg and was also within normal limits. (Table 9)

In addition,intra operative data related to On-pump CABG revealed that the mean duration of usage of CPB pump was 98.23 ± 9.91 minutes whereas the mean duration of Aortic Cross Clamp was 60.20 ± 4.31 minutes.The entire study participants (N=44) received Antegrade Cardioplegia . About 14% (n=6) required IABP support to maintain stable hemodynamics.The number of grafts used on an average was 3± 0.81.(Table 9)

Outcomes:

Our primary outcome was to evaluate if the preoperative level of the cardiac biomarker NT-Pro BNP, correlates with intraoperatively measured Pulmonary Capillary Wedge Pressure (PCWP) in patients undergoing On-Pump CABG surgery.

- **Cardiac biomarker, NT-Pro BNP and PCWP:**

Table 10: Linear relationship of Pre-operative NT-Pro BNP with Intraoperative PCWP

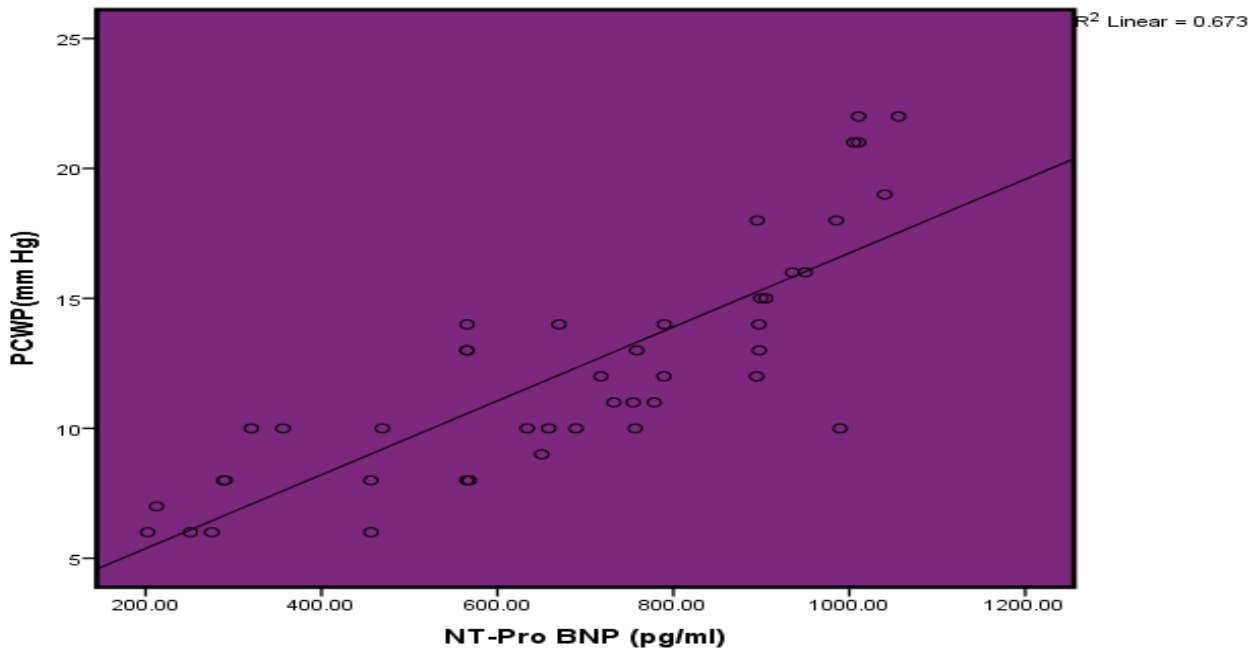
		PCWP(mm Hg)
NT-Pro BNP (pg/ml)	r-value	.821**
	P-value	.000

**P<0.01; *P<0.05

The linear relationship of pre-operative NT-Pro BNP with PCWP was analyzed using Pearson's correlation Analysis.It was inferred that a strong positive linear relationship existed between NT-Pro BNP level and PCWP level with an r value of 0.821** and a p value <0.01.This indicates that when preoperative NT-Pro BNP levels increase , intraoperative PCWP levelsalso increases

Figure 18: Scatter plot for correlation of Preoperative NT-Pro BNP with Intra-operative

PCWP



- **Inflammation and Dobutamine Concentration :**

The normality of TNF Alpha data obtained both pre and intraoperatively were assessed using Shapiro-wilk test and found that the data followed normality assumptions (p >0.05).(Table 11)Box-whisker plot for the baseline TNFα and intra-operative TNFα depicted are depicted in a graphical manner (Figure 19(a)& (b)). Hence, paired sample t test, a parametric statistical test was used to compare the preoperative TNFα and intra-operative TNFα levels.

Table 11: Tests of Normality for TNFα data

	Shapiro-Wilk Statistic	df	P-value
Pre-operative TNF-Alpha(pg/ml)	.959	44	.116
Intraoperative TNF-Alpha(pg/ml)	.951	44	.058

Figure 19: Box & Whisker Plots for (a) Preoperative TNF-Alpha levels
(b) Intraoperative TNF-Alpha Levels

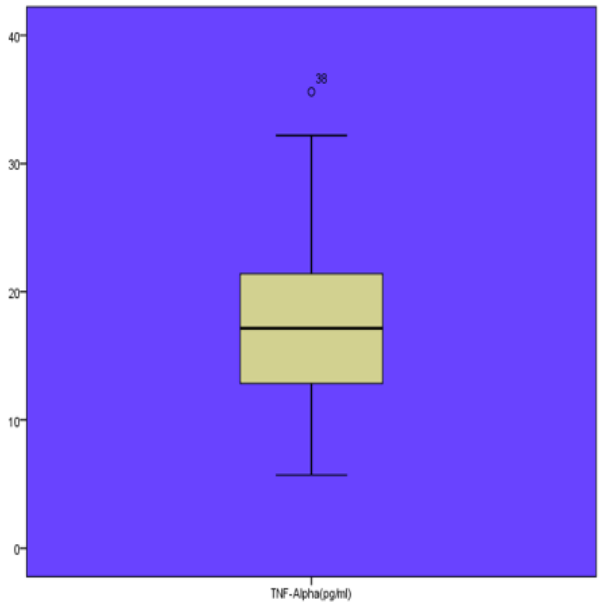


Figure 19 (a)

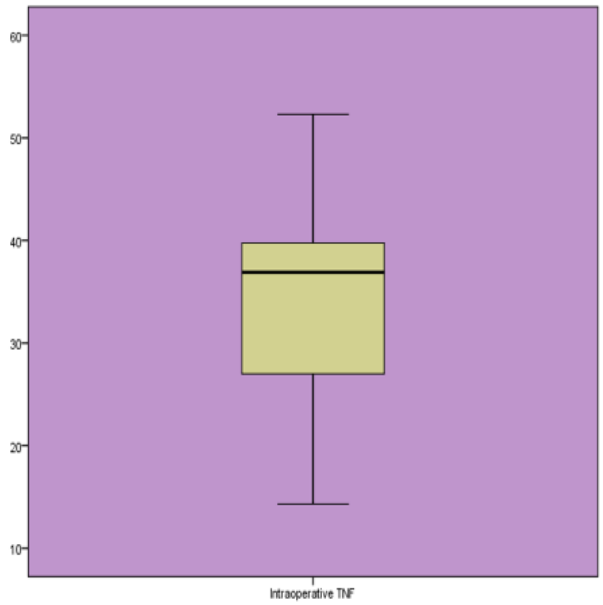


Figure 19 (b)

Table 12: Comparison of TNFα levels in the preoperative and intra-operative periods

	Baseline	Intraoperative	t-value	P-value
	Mean±SD			
TNF-Alpha(pg/ml)	17.92±7.04	33.75±9.65	-18.307**	0.001

**P<0.01; *P<0.05

The mean preoperative levels of TNF-Alpha (17.92±7.04 pg/ml) falls within the normal levels whereas the mean intraoperative levels of TNF-alpha (33.75±9.65) were abnormally elevated. The use of paired sample t test revealed that there was a statistically highly significant increase in the level of TNFα during intra-operative state compared with the preoperative state for a level of significance set at 1% (P<0.01). (Table 12)

Table 13: Linear relationship between Intraoperative TNFα and Plasma Dobutamine Concentration

		Plasma Dobutamine Steady State ConcentrationCPss (mcg/ml)
TNF-Alpha (pg/ml)	r-value	.081
	P-value	.601

**P<0.01; *P<0.05

Pearson correlation analysis was done to determine the relationship ofintraoperativelevels of TNFα with Plasma DobutamineSteady State Concentration (CPss)for a 5% level of significance. It was inferred that there was no statistically significant relationship between TNFα and Plasma Dobutamine concentration at steady state with p>0.05. (Table 13 and Figure 20)

Figure 20: Scatter plot demonstrating relationship of Intraoperative TNFα and Plasma Dobutamine Steady State Concentration (CPss)

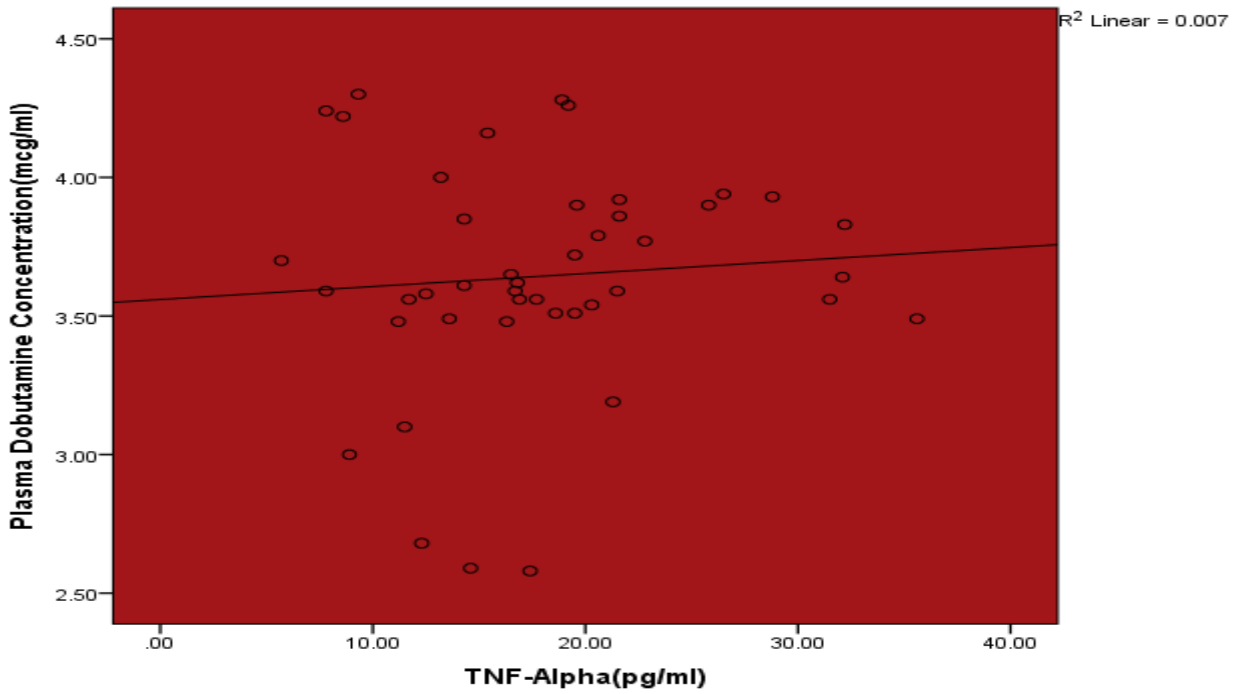


Table 14: Relationship between Dobutamine Dose and Plasma Dobutamine Steady State Concentration (CPss)

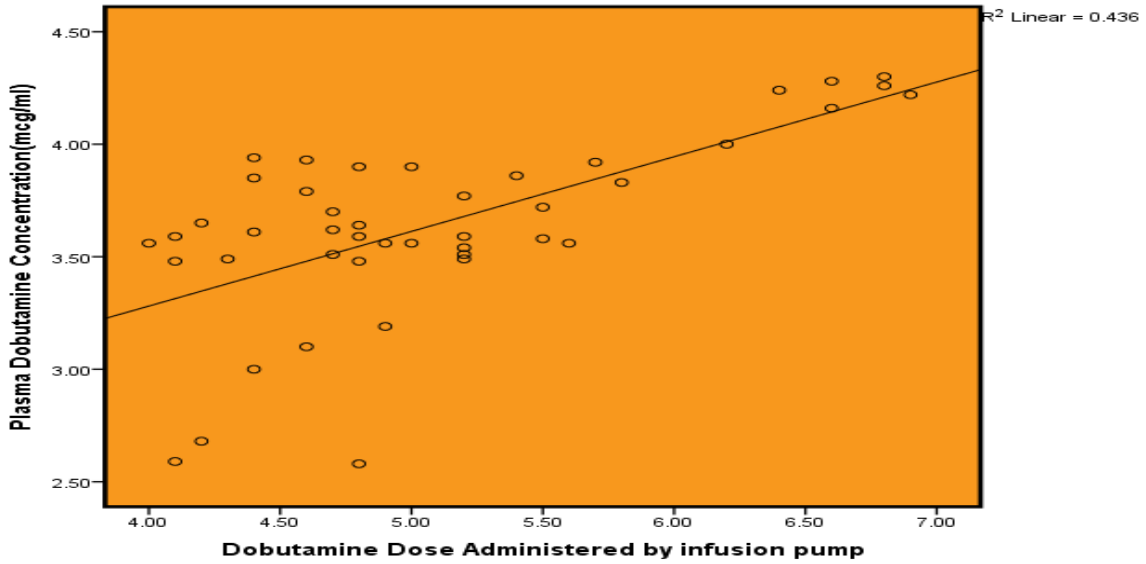
		Dobutamine Dose Administered by infusion pump	Plasma DobutamineSteady-StateConcentration (CPss) (mcg/ml)
DobutamineDose Administered by infusion pump	r-value	1	.660**
	P-value		.000

**P<0.01; *P<0.05

From the above table (Table 14), it was observed that dobutamine doseadministered by infusion pump and dobutamine concentration in plasma at steady state

(Cpss)exhibited a positive linear relationship with one another with p<0.01.Hence, when Dobutamine dose levels administered by infusion pump increases, plasma dobutamine concentration will also increase and vice-versa. (Figure 18)

Figure 21: Scatter plot demonstrating relationship of Plasma Dobutamine concentration at steady state (CPss) with the administered Dobutamine dose



- **Biomarkers andCo- morbidities**

Our tertiary outcome was to evaluate the role played by NT-pro BNP and TNF-α in the pre-existing co-morbidities.For this we compared the preoperative levels of NT-pro BNP and TNF-α among those with and without the comorbidities. By performing independent sample t test it was concluded that there was no difference in the level of NT Pro BNP and TNFα (P>0.05) among participants with and without the co-morbidities.(Table)

Table 15: Comparison of the level of NT-Pro BNP and TNF-Alpha level among individuals with and without various co-morbidities

	Hypertension		**t-value**	**P-value**
	Yes	**No**		
NT-Pro BNP (pg/ml)	676.41±271.93	725.68±171.61	0.489	0.628
TNF-Alpha(pg/ml)	17.91±7.30	17.96±6.13	0.019	0.985
	DM		**t-value**	**P-value**
	Yes	**No**		
NT-Pro BNP (pg/ml)	678.04±255.84	707.36±266.18	0.326	0.746
TNF-Alpha(pg/ml)	17.69±7.17	18.61±6.90	0.371	0.713
	Thyroid		**t-value**	**P-value**
	Hypothyroid	**No**		
NT-Pro BNP (pg/ml)	754.26±187.63	667.65±269.79	-0.905	0.371
TNF-Alpha(pg/ml)	15.81±7.73	18.46±6.86	1.008	0.319
	Dyslipidemia		**t-value**	**P-value**
	Yes	**No**		
NT-Pro BNP (pg/ml)	694.62±251.98	613.17±303.34	-0.666	0.509
TNF-Alpha(pg/ml)	17.75±7.37	19.28±3.79	0.454	0.652
	H/o Previous MI		**t-value**	**P-value**
	Yes	**No**		
NT-Pro BNP (pg/ml)	602.23±311.00	696.03±250.49	0.769	0.446
TNF-Alpha(pg/ml)	21.56±6.31	17.45±7.06	-1.235	0.224
	H/o Stroke		**t-value**	**P-value**
	Yes	**No**		
NT-Pro BNP (pg/ml)	743.36±137.65	679.57±264.99	-0.472	0.640
TNF-Alpha(pg/ml)	12.65±6.55	18.45±6.94	1.599	0.117

Figure 22: Elevated levels of NT-Pro BNP and Pre-Existing Co-morbidities:

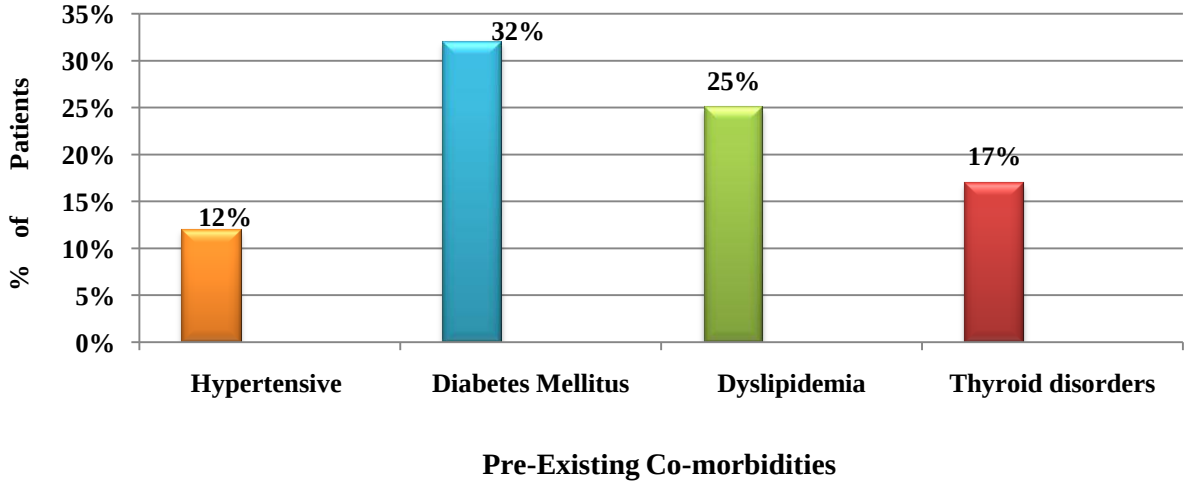
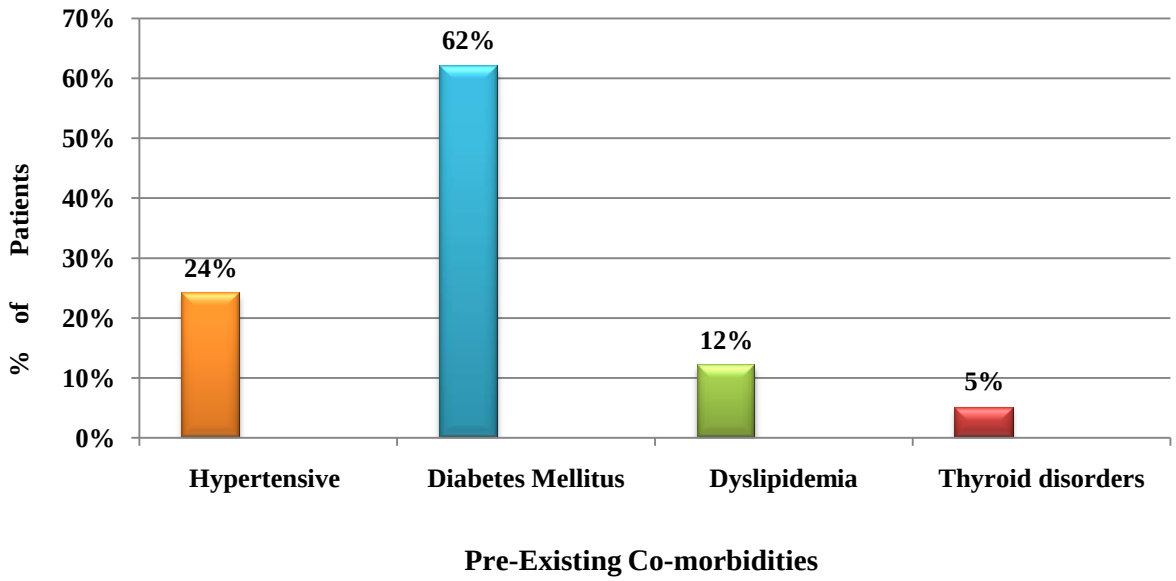


Figure 23: Elevated levels of TNF-Alpha and Pre-Existing Co-morbidities:



Among individuals with increased levels of NT-Pro BNP, it was observed that 12% were hypertensives, 32% were Diabetics, 25% of individuals had dyslipidemia and 17% of individuals had thyroid dysfunction (Figure 22).

Among individuals with increased levels of TNFα, it was observed that 24% were hypertensives , 62 % were Diabetics, 12% of individuals haddyslipidemia and 5% of individuals had thyroid dysfunction (Figure 23).

Comparison of Pre- operative NT-Pro BNP and TNF-Alpha level among male and female patients

Table 16: Tests of Normality for gender wise NT-Pro BNP and TNFα

	Sex	Shapiro-Wilk Statistic	df	P-value
Preoperative NT-Pro BNP (pg/ml)	Male	.936	28	.089
	Female	.936	16	.301
Pre- operative TNF-Alpha(pg/ml)	Male	.959	28	.331
	Female	.959	16	.652

For 5% level of significance, NT-Pro BNP and TNFα levels of male and female patients satisfied the normality assumptions (P>0.05) completely, hence we infer that Preoperative levels of NT-Pro BNP and TNFα amongboth male and female patients follows normal distribution.(Table 16) This is represented graphically by Box-whisker plot (Figure 24 (a)&(b)). Therefore, parametric statistical test (unpaired t-test)can be used to compare the Preoperative levels of NT-Pro BNP and TNFα between male and female patients.

Using independent sample t test, for level of significance set at 5% it is inferred that male and female patients had no statistically significant differences (P>0.05) amongthem with respect to pre-operative values ofNT-Pro BNP and TNF-Alpha.(Table 17)

Figure24 :Preoperative levels of NT-Pro BNP and TNF-alpha among (a) males and (b) females

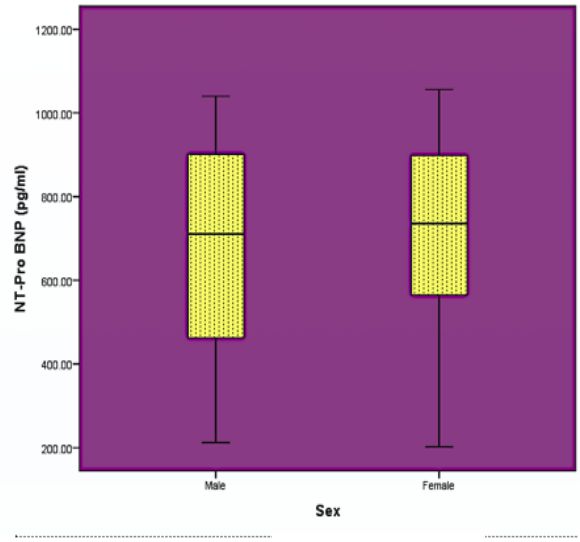


Figure 22 (a)

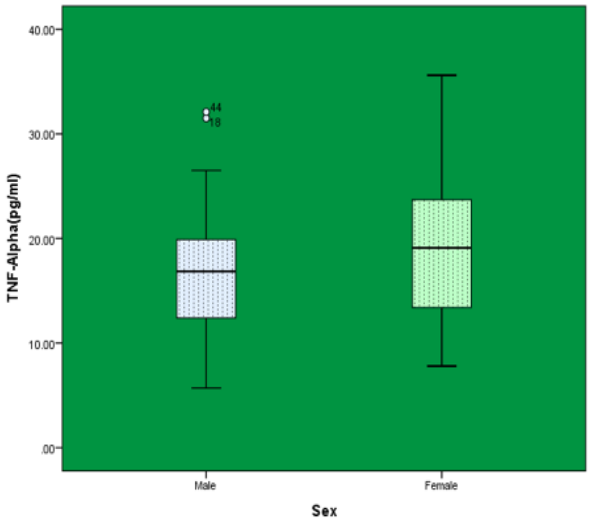


Figure 22 (b)

Table 17: Comparison of NT-Pro BNP and TNF-Alpha levels between male and female patients

	Male	**Female**	**t-value**	**P-value**
	Mean± SD			
NT-Pro BNP (pg/ml)	678.10±254.17	698.07±266.07	-0.247	0.806
TNF-Alpha (pg/ml)	17.13±6.31	19.31±8.19	-0.991	0.327

Using independent sample t test, for level of significance set at 5% it is inferred that male and female patients had no statistically significant differences (P>0.05) amongthem with respect to pre-operative values ofNT-Pro BNP and TNF-Alpha.(table)

Comparison of Preoperative NT-Pro BNP and TNF-Alpha level among various age group patients

Table 18: Tests of Normality for age group wisePreoperative NT-Pro BNP and TNF-Alpha data

	Age group	Shapiro-Wilk Statistic	df	P-value
NT-Pro BNP (pg/ml)	41-50	.904	11	.207
	51-60	.929	13	.331
	61-70	.939	20	.231
TNF-Alpha(pg/ml)	41-50	.884	11	.118
	51-60	.918	13	.239
	61-70	.932	20	.168

Normality verification test clearly indicated that NT-Pro BNP and TNFα data of all age group patients followed normal distribution (P>0.05). Hence, parametric statistical test Analysis of Variance (ANOVA) was used to compare the NT-Pro BNP and TNF-Alpha levels among various age group patients.(Table 18). The box-whisker plot (Figure 25a&b) graphically represent the normality assumptions.

Figure 25: Age group wise Preoperative (a) NT-Pro BNP and (b) TNF-Alpha data

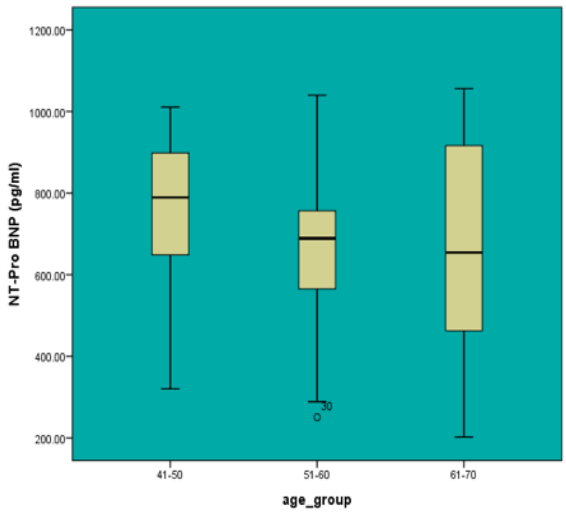


Figure 23 (a)

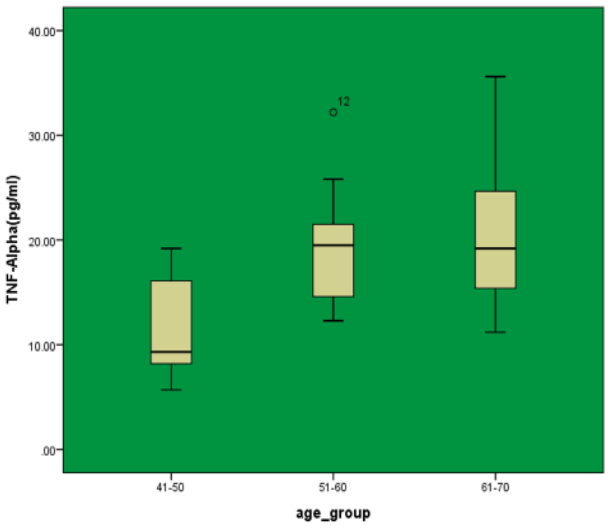


Figure 23 (b)

Table 19:Comparison of Preoperative NT-Pro BNP and TNF-Alpha levels among various age group patients

		Mean±SD	F-value	P-value
NT-Pro BNP (pg/ml)	41-50	757.15±222.62	0.575	0.567
	51-60	654.36±259.69		
	61-70	666.04±274.22		
TNF-Alpha(pg/ml)	41-50	12.04±4.88	6.671**	0.003
	51-60	19.03±5.59		
	61-70	20.44±7.23		

**P<0.01; P<0.05

Comparison of Preoperative NT-Pro BNP and TNF-Alpha levels among various age group patients was performed using Analysis Of Variance (ANOVA) test. Analysis

ofNT-Pro BNP values forlevel of significancesetat 5%, showed that there was no statistically significant difference among various age group patients.

However,analysis of TNF-Alpha values for level of significance setat 5%a highly statistically significant difference (P<0.01) was noticed in the TNF-Alpha level among different age group patients. It was identified that individuals in the age group of 41-50 years low levels of TNFα compared with patients of other age groups. (Table 19)

Table 20: Relationship between various intraoperative hemodynamic parameters

		Heart Rate(beats per minute)	SYSTOLIC	DIASTOLIC	PCWP(mm Hg)	CVP (mm Hg)
Heart Rate(beats per minute)	r-value	1	-.165	-.026	.108	-0.062
	P-value		.286	.866	.487	0.690
SYSTOLIC	r-value		1	.873**	-.099	0.023
	P-value			.000	.523	0.880
DIASTOLIC	r-value			1	-.009	-0.024
	P-value				0.951	0.878
PCWP (mm Hg)	r-value				1	0.167
	P-value					0.279
CVP (mm Hg)	r-value					1
	P-value					-

Pearson correlation analysis showed that there was no correlation of PCWP with hemodynamic parameters like heart rate, systolic and diastolic BP. Similarly, there was no correlation of CVP with hemodynamic parameters like heart rate, systolic and diastolic BP. However, systolic and diastolic blood pressures were found toexhibit a linear relationship with one another.(Table 20)

Table 21: Relationship between NT-Pro BNP, Intraoperative TNF-Alpha, Aortic Cross Clamp Duration and CPB Pump Duration

		NT-Pro BNP (pg/ml)	Intraoperative TNF	Aortic Cross Clamp Duration (mins)	CPB Pump Duration (mins)
NT-Pro BNP (pg/ml)	r-value	1	-.249	.045	-.069
	P-value		.103	.769	.654
Intraoperative TNF (pg/ml)	r-value		1	-.050	-.071
	P-value			.746	.647
Aortic Cross Clamp Duration (mins)	r-value			1	.401**
	P-value				.007
CPB Pump Duration (mins)	r-value				1
	P-value				-

**P<0.01; P<0.05

It was inferred from Pearson correlation analysis that CPB pump duration and Aortic cross clamp duration were positively correlated with one another (P<0.01).This is graphically represented in the below scatter plot.Where aspre-operative NT-Pro BNP and Intraoperative TNF- alpha levels did not show any correlation with CPB pump duration and Aortic cross clamp duration. (Table 21)

DISCUSSION

Biomarkers are well acknowledged for risk prediction and stratification in patients with Coronary Artery Disease (CAD) however their value in the perioperative period among patients undergoing On-Pump CABG surgery, which is the gold-standard surgical revascularization procedure is less explored. If identified to be valuable diagnostic or prognostic tools they can influence decision-making significantly hence leading to better patient outcomes.

In this study, we investigated the prognostic role and correlation of a cardiac biomarker, NT-Pro BNP with PCWP, an important hemodynamic parameterreflecting the development of Low Cardiac Output Syndrome (LCOS) and the role of an inflammatory biomarker TNF-Alpha, released as a part of SIRS resulting from the use of CPB pump on the plasma concentration of the inotrope dobutamine that is used to combat the LCOS.

The Cardiac biomarker, NT-Pro BNP and hemodynamic parameter , PCWP in patients undergoing On-Pump CABG surgery

According to the ESC guidelines, for the diagnosis and treatment of acute and chronic Heart failure (2016), in a non-acute setting the upper limit of normal for NT-proBNP is 125 pg/mL, while in an acute setting, the cut-off value for NT-proBNP is 300 pg/mL.[75] In this study with reference to the ESC 2016 Guidelines , it was observed that the preoperative levels of NT-Pro BNP were elevated among the study

participants with a mean of 685.37±255.64 pg/ml (Table 6). This elevation in preoperative NT-Pro BNP levels could be explained to be due to the reduced LV function in patients of CAD who require CABG as a procedure of revascularization. As per numerous studies NT-Pro BNPhas been shown to be an independent predictor of adverse CV events in patients with stable CAD.[128-130]

Based on the ICON study, when age-dependent cut-offs of NT-pro BNP are taken into consideration it was observed that the levels were predominantly elevated in individuals belonging to the age group of 41-50 years with mean NT-Pro BNP levels of 757.15±222.62pg/ml (Table 19) while the acceptable cutoff is 450 pg/ml.[82] However statistical analysis of NT-Pro BNP levels among different age groups using ANOVA did not show any statistically significant difference between 41-50 years age group with other age groups (p=0.567).(Table 19)

It is well established that blood levels of BNP and NT Pro BNP are elevated following LV systolic dysfunction wherein, increased ventricular filling pressure increases blood levels of BNP. Other conditions where in NT-Pro BNP levels may be increased in addition to LV systolic dysfunction include renal failure and female gender .[96,97]In this study, subjects with renal failure (Serum creatinine >1.6)were excluded during screening in order to avoid confounding.

Redfield et al., concluded that NT-Pro BNP levels were significantly higher among females than males hence interpretation of NT-Pro BNP values should be gender

specific.[98] However, in this study population the mean NT-Pro BNP among males and females were 678.10±254.17pg/ml and 698.07±266.07pg/ml respectively. Further, analysis using independent sample t test showed no statistically significant difference of NT-Pro BNP based on gender (p=0.806). (Table 17) This indifference of the present study with the preexisting study results could likely be due to the reason that this study population did not have an equal distribution of males and females.

According to various studies, plasma NT- proBNP is considered as a useful marker of recovery after a high-risk CABG surgery, and significantly correlated in an inverse manner with LVEF i.e NT-Pro BNP levels increases as the Left Ventricular Ejection Fraction(LVEF) declines.LV systolic dysfunction in LCOS leads to reduction in LVEF and increase in LV filling pressure and this is closely associated with high NT-proBNP levels .[99,100,104]Studies have also demonstrated that whenever LVEF declines as a result of LCOS, PCWP increases.[64,65,68]

In the current study, the mean PCWP of all 44 participants was found to be 12.27±4.43 mm of Hg, hence on average the study population had a PCWP on the higher side of normal. (Table 9) When patients are monitored using a pulmonary artery catheter , congestion corresponds to elevated pulmonary capillary wedge pressure (PCWP), generally >16 mm of Hg. It has also been shown that even at levels below symptom threshold, elevated PCWP predicts worse outcome in heart failure patients. [95]

The primary outcome analysis of this study included the correlation of preoperatively measured NT-Pro BNP values with intraoperatively measured PCWP. For all the

subjects (N=44), their baseline NT-Pro BNP levels and intraoperatively measured mean PCWPwere subjected to Pearson's correlation analysis, from which it was inferred that a strongly positive linear relationship existed between NT-Pro BNP and PCWP with an r value of 0.821^{**} and a p value $<0.01^{**}$.(Table10 ,Figure 18) Hence,the trend was such that whenever preoperative NT-Pro BNP levels increases there was an increase in intraoperative mean PCWP levels also. In other words, it was established that a positive correlation exists between NT-Pro BNP and PCWP.

Hence, this research provides a valid information that pre-operativeNT-Pro BNP when increased more than 300pg/ml indicates the need for Cardio Pulmonary Bypass pump with the inotrope Dobutamine subjected tofrequent dose adjustments while performing CABG procedure in order to avoid development of LCOS on table.

Although to the best of our knowledge there are no correlation studies of NT-Pro BNP with PCWP in the setting of on-pump CABG surgery till date, there do exists correlation studies for the above parameters in other settings. A study conducted by Forfia et al., prospectively investigated if NT-Pro B-type natriuretic peptide could serve as a non-invasive marker of pulmonary capillary wedge pressure (PCWP) in 40 ICU patients requiring invasive hemodynamic monitoring. They concluded that the natriuretic peptide levels were markedly elevated and correlated with invasive hemodynamic parameters, most notably PCWP.[101]This finding was consistent with results of the current study.

Similar to the primary outcome findings of this study, another two prospective studies conducted by Kazanegra et al.,and Cheng et al., in patients with decompensated heart

failure demonstrated a strong positive correlation between NT-Pro BNP and PCWP.[102,103]However in contrast to the findings of our current study, a few prospective studies have also demonstrated a negative correlation between NT-Pro BNP and PCWP in the ICU setting.[104-106]This discordance can be explained on the basis of the different setting in which this correlation study was conducted.

Role of the inflammatory marker TNF-Alpha on the Plasma Concentration of Dobutamine in patients undergoing On-Pump CABG surgery

In this current study ,it was inferred that in comparison to TNF-Alpha values measured preoperatively (mean Preoperative TNF-Alpha -17.92 ±7.04pg/ml), the values were elevated when TNF-Alpha was measured from samples collected intraoperatively at 4^{th} hour from incision(mean Intraoperative TNF-Alpha -33.75±9.65pg/ml). Analysis using Paired t test was done to determine if the difference in the intraoperative period was statistically significant in comparison to pre-operative period. It was concluded that the increase in TNF-alpha levels in the intraoperative period in comparison to the preoperative period was highly statistically significant ($P<0.01$). (Table 12)This increase in the inflammatory marker during surgery was attributed to the use of the cardiopulmonary bypass pump (CPB)which as per various studies elicits a SIRS leading to a increase in various pro-inflammatory cytokines including TNF-α. [83,86,87-89]

As per this study, the finding of elevated TNF-Alpha in the intraoperative period was found to be in concordance with the findings of El Azab et al who investigated the

cytokine response between patients undergoing CABG surgery with CPB (On-Pump) and without CPB pump(Off-Pump). They had concluded that TNF-Alpha levels among patients undergoing On-Pump CABG surgery was significantly higher in comparison to those undergoing Off-Pump CABG surgery and they ascribed this difference to the extracorporeal circulation by the CPB pumps inducing a marked inflammatory response and resulting in deleterious systemic effects.[107]

Similarly, numerous other studies performed using both clinical CPB and pre-clinical models of CPB have found that the plasma levels of the proinflammatory cytokine TNF-α is significantly increased during and following CPB.[108-111] Zhang et al., identified that the plasma level of TNF-α in patients undergoing on-pump CABG surgery significantly increases when CPB begins and reaches a peak following CPB.[109]According to Bittar et al., it was concluded that the peak in TNF-Alpha generally occurs around the 4th hour from incision.[91]This time point was followed in our study for sample collection in order to analyse TNF-Alpha levels.

In contrast to the findings of this study, Welters et al.,and Martinez-Comendador et al., reported that plasma TNF-α levelsare not statistically different pre- and post-surgery. The discrepancy may be due to the differences in thestudy design, procedure as well as the complex chemistry of TNF-α during analysis.[112,113]

In the current study , the presence of a linear relationship between Plasma dobutamine concentration and TNF-Alpha levels that were obtained at 4th hour from incision, was tested using Pearson’s correlation analysis. The results revealed that there was no

statistically significant relationship between the intraoperative levels of TNFα and Plasma Dobutamine concentration (r=0.81 and p>0.05).(Table 13,Figure 20) Hence it was found that increase in TNF-Alpha levels following the use of CPB does not significantly interfere with the steady state concentration of Dobutamine, the inotropic drug that is most commonly used to combat LCOS and maintain adequate cardiac output in the setting of CABG.This may be explained on the basis of 100% bioavailability achieved with IV administration that hence indicates clinical management of LCOS using the inotrope Dobutamine is effective in spite of inflammation.

The present study is the first to examine the role of TNF-Alpha on the plasma concentration of the frequently used inotropic drug – dobutamine, in patients undergoing on-pump CABG surgery. The few correlation studies of TNF-Alpha with Dobutamine that exists previously were performed in different settings.

Hartemink KJ et al., prospectively studied 20 patients admitted to the ICU for treatment of septic shock. Their study findings revealed the presence of a positive association and relation between TNF-α and dobutamine concentrations. [114]

Whereas , Sun D et al., reported that the correlation between dobutamine concentration and TNF-Alpha levels are dobutamine dose dependent in a setting of unilateral total hip arthroplastyin elderly patients . Low-dose dobutamine (2 μg/kg/minor 4 μg/kg/min) exhibited a negative correlation with TNF-Alpha levels.However, high-dose dobutamine hydrochloride (6 μg/ kg/min) exhibited no significant correlation with plasma TNF-α level. [115]However as these correlation

studies of TNF-alpha with Dobutamine levels are performed in different settings they may not be comparable.

This study findings also revealed that the dose of dobutamine administered by infusion pump and plasma dobutamine concentration attained at steady state (CPss) had ahighly significant positive linear relationship with one another ($p < 0.01$). Increase in dobutamine dose was associated with a concomitant increase in dobutamine steady state concentration. (Table 14 and Figure 21)

This finding of this study is in concurrence with the findings of Leier et al.who concluded that a positive linear relationship exists between dobutamine dose and the resulting plasma concentration in adults.[56] Similarly Mahoney et al., also reported that dobutamine concentrations were positively correlated with infusion dosages, however this study was performed among neonatal age group. It was also stated that the range of values shows wide interindividual variations despite similar doses.[116]

Role of Cardiac biomarker (NT Pro BNP) and Inflammatory marker (TNF-Alpha)on pre-existing co-morbidities

The current study also investigated the role played by NT-pro BNP and TNF-α with regard to pre-existing co-morbidities in patients undergoing on-pump CABG surgery. For this evaluation , the basal levels of NT-pro BNP and TNF-α among those with and without comorbidities were compared using independent sample t test and was inferred that there was no difference in the level of NT Pro BNP and TNFα ($P>0.05$)

among participants with and without the co-morbidities.(Table 15) Hence, according to this study, the cardiac biomarker NT-Pro BNP and the inflammatory marker, TNF-Alpha were not significantly altered in patients with various comorbidities included in our study such as Hypertension,DiabetesMellitus,Hypothyroidism and Dyslipidemia.

This study also revealed that increased levels of NT-Pro BNP was observed among 12% of hypertensives, 32% of Diabetics, 25% of individuals with dyslipidaemia and 17% of individuals with thyroid dysfunction (Figure 22). In addition, increased levels of TNFα was observed among 24% of hypertensives, 62 % of Diabetics, 12% of individuals with dyslipidemia and 5% of individuals with thyroid dysfunction (Figure 23).

Evidence on the association of NT-Pro BNP with hypertension is mixed. Findings of few studies were in contrast to the results of this study. In a cross-sectional study conducted among 202 participants with history of dyspnoea, the mean NT-proBNP levels were estimated to be 60% higher in individuals with diagnosed hypertension compared to those without hypertension. A case-control study among 48 African patients with hypertension and 20 normotensive participants inferred that the mean NT-proBNP concentration was around 20 times higher in the hypertensive group.[117,118]These findings were in discordance to our study. This could be probably because the significance of association in this study was tested for a relatively smaller sample size and without adequate follow up.

In disagreement to the findings of the present study regarding the absence of association of NT-Pro BNP with Diabetes Mellitus and Dyslipidaemia, studies done previously by Lazo et al., Neeland et al., and Schlueter et al., have reported that increased NT-Pro BNP has been found to be inversely associated with metabolic disorders such as diabetes , dyslipidemia because of its postulated role in adipose and glucose metabolism.A study was conducted by Van de Horst et al., on 371 patients with heart failure, 81 of whom had diabetes.The patients were monitored for five years.It was concluded thatamong patients with diabetesNT-pro-BNP levels were significantly higher than in those who did not survive. [119-121,131]The absence of significance in this study could be due to the fact that patients who are posted for CABG surgery are well controlled for diabetes prior to surgery. In addition differences in the study population could also account for the difference of the current study as not all the participants of the present study had pre-existing heart failure as with the study in comparison.

In contrast to the findings observed in this study that TNF-Alpha levels are not significantly different among hypertensives and non-hypertensives,Mohamed et al., concluded that TNF-alpha levels are remarkably increased in hypertensive patients and may also play an important role in the pathogenesis and development of renal damage in hypertensive patients. Also a recent study by Kroetsch and his colleagues demonstrated that TNF uses an unconventional mechanism that regulates blood vessel constriction. Instead of activating a conventional "forward signal" through TNF receptors, TNF in the vascular wall initiates a "reverse signal" that directly travels into the contractile smooth muscle cells. [122,123]

Again in contrast to the result that TNF alpha levels are not significantly different between diabetics and non-diabetics in the present study population, various studies have found a significant increase in TNF-Alpha including a recent study by Akash et al., that concluded that TNF-Alpha is one of the most important proinflammatory cytokine that participates in the pathogenesis of DM and is significantly elevated in such patients. [124-127]This discrepancy of the results of this current study that TNF-Alpha levels are not elevated in Diabetic patients could be due to the reason that their blood sugar levels are brought well under control prior to surgery, hence unlike in comparison to uncontrolled diabetics their level of inflammation does not prove to be significant.

This study suggests that patients with Diabetes alone or in combination with Systemic hypertension/Hypothyroidism/ Dyslipidaemia may requireTNFα estimation prior to the end of CABG procedure in order to avoid post of complication like ischemia – reperfusion injury , low output state , organ dysfunction. [90]

Strengths of this study:

i. Novelty of the research question of the present study as no studies have been done till date in a similar setting of patients undergoing On-Pump CABG surgery.
ii. This study assessed multiple variables for multiple outcomes in a sample size that was just adequate for a first time study. It thereby helped us draw

conclusions that would help in the development of future large scale research for relevant exposure and outcome variables.

iii. Various confounders that would interfere with the study outcome were controlled by excluding such participants from the study before recruitment.

iv. This study used validated procedures for estimation of NT-Pro BNP, TNF Alpha using ELISA and Dobutamine by HPLC .

Limitations of our Study:

i. This study was performed among a population of patients attending a single centre. Although sample size calculation was performed based on existing literature, it was relativelysmall mainly because, this study was designed for critical care patients (CABG). It may limit the generalizability of our findings to the entire population.

ii. This study was performed only among patients undergoing On-Pump surgery electively. Inclusion of patients undergoing emergency surgery would provide a broader perspective.

iii. Blinding was not performed for the principal investigator, the surgeons, the anaesthetist and the paramedical staff due to the procedures involved in the critical care management setup. This could have resulted in bias in the assessment of Pulmonary capillary wedge pressure.

CONCLUSION

This is the first study conducted among On-Pump CABG patients to investigate the role of a cardiac biomarker NT-Pro BNP and an inflammatory marker TNF-α on the Pulmonary Capillary Wedge Pressure (PCWP) and its effect on the plasma concentration of the inotrope Dobutamine used during CABG procedures .

This study revealed the presence of a statistically significant, strongly positive correlation between preoperative NT-Pro BNP values and intraoperatively measured PCWP. Hence, we conclude that in an acute setting when pre-operativeNT-Pro BNP levels are increased more than 300pg/ml, the need of cardio pulmonary bypass pump withfrequent dose adjustments and monitoring of the inotrope Dobutamine may be necessary to avoid the development of LCOS on table.

Based on the findings in this study, Preoperative NT-proBNP could serve as valuable marker in predicting intraoperative risk of developing LCOS in patients undergoing on-pump CABG surgery and aid anaesthetists and cardiothoracic surgeons in decision making regarding patients admitted at a critical care setup.

The study demonstrated a statistically significant increase in TNF-Alpha levels in the intraoperative period in comparison to the preoperative period which is in concordance to numerous studies and can be attributed to the use of CPB pump.

The present study also revealed the presence of a statistically significant linear relationship between dobutamine dose administered by infusion and the plasma concentration of dobutamine at steady state.This may be attributed to 100% bioavailability on IV administration and hence indicates that inotropic support with dobutamine using an infusion pump serves to be an effective management strategy in the prevention of LCOS on table .

However, this study found that there is no statistically significant linear relationship between intraoperative TNF-Alpha values and plasma dobutamine concentration at steady state which along with linear relationship exhibited by dobutamine dose administered by infusion pump with plasma concentration of dobutamine help us conclude that intraoperative monitoring of TNF-Alpha during surgery may not be necessary.

In addition this study results revealed the absence of any significant association between the biomarkers (NT-Pro BNP & TNF-Alpha) and pre-existing co morbidities among the study population which was concluded to be due to these co-morbidities being well controlled prior to surgery. It was also inferred that in Patients with Diabetes alone or in combination with Systemic hypertension/ hypothyroidism/ dyslipidaemia TNFα estimation prior to the closure of CABG surgery could avoid post-operative complications like ischemia –reperfusion injury , low cardiac output syndrome , organ damage.

Therefore, preoperative NT-Pro BNP estimation may help prevent the development of LCOS on table and estimation of TNF-Alpha prior to the end of surgery may aid in prevention of adverse post-operative outcomes.

Future prospective studies with larger sample size and randomized trial designs could further expand the knowledge about these biomarkers(NT-Pro BNP & TNF-Alpha) among patients undergoing On-Pump CABG surgery and may help improve patient outcome in the post-operative period.

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ABBREVIATIONS:

ACC- Aortic Cross Clamp

ACCF/AHA - American College of Cardiology Foundation/American Heart Association

AHF- Acute Heart Failure

AMI- Acute Myocardial Infarction

ANOVA- Analysis Of Variance

ATP- Adenosine Triphosphate

BMI- Body Mass Index

BNP- B-type natriuretic peptide

BSA- Body Surface Area

CABG -Coronary Artery Bypass Grafting

CAD- Coronary Artery Disease

cAMP- cyclic Adenosine Mono Phosphate

CHD -Coronary Heart Disease

CI- Cardiac index

CK-MB – Creatinine Kinase-MB

CO-Cardiac Output

COMT- Catechol-O-Methyltransferase

CPB- Cardio-Pulmonary Bypass

CPss- Steady State Plasma Concentration

CVD - Cardiovascular Disease

CVP- Central Venous Pressure

EDTA- Ethylene Diamine Tetra Acetic acid

EF- Ejection Fraction

ELISA- Enzyme-Linked Immunosorbent Assay

ESC- European Society of Cardiology

ESV- End Systolic Volume

FDA- Food and Drug Administration

HPLC-High Performance Liquid Chromatography

ICON- International Collaborative of NT-pro BNP

ICU- Intensive Care Unit

LAD- Left Anterior Descending

LAP-Left Atrial Pressure

LCOS- Low Cardiac Output Syndrome

LIMA- Left Internal Mammary Artery

LoD- Limit of Detection

LoQ- Limit of Quantification

LVEDP- Left Ventricular End-Diastolic Pressure

LVEF- Left Ventricular Ejection Fraction

LVF- Left Ventricular Function

LVSD- Left Ventricular Systolic Dysfunction

MI- Myocardial Infarction

NCDs - Non Communicable Diseases

NEP-Neutral Endo Peptidase

NNT- Number Needed to Treat

NPR-C - Natriuretic Peptide Receptor-C

NT-pro BNP- N-terminal pro BNP

PAC - Pulmonary Artery Catheter

PAP- Pulmonary Artery Pressure

PCI- Percutaneous Coronary Intervention

PCWP- Pulmonary Capillary Wedge Pressure

PVR-Peripheral Vascular Resistance

RCT- Randomized Controlled Trials

RIJV- Right Internal Jugular Vein

RWMA- Regional Wall Motion Abnormality

SIRS- Systemic Inflammatory Response Syndrome

SVC-Superior Vena Cava

SYNTAX Trial - Synergy between PCI with Taxus and Cardiac Surgery Trial

TEE- Transoesophageal echocardiography

TGL- Triglycerides

TNF-α -Tumour Necrosis Factor-α

TVD- Triple Vessel Disease

PSG Institute of Medical Science and Research, Coimbatore
Institutional Human Ethics Committee
INFORMED CONSENT FOR RESEARCH PROJECTS

I **Dr.R.Subhashini** am carrying out a study on the topic**:A Study To Evaluate The Association Of Biomarkers With Pulmonary Capillary Wedge Pressure And Plasma Concentration Of The Inotrope Dobutamine In Patients Undergoing On-Pump CABG Surgery In A Super-Speciality Hospital At Coimbatore : A Prospective Observational Study**as part of my research project being carried out under the aegis of the Department of Pharmacology.

My research guide is**: Prof. Dr. K. Bhuvaneswari**

The justification for this studyis:

- Based on Pubmed search ,no study on the correlation of Preoperative NT-pro BNP with PCWP in patients undergoing On-Pump CABG in India.
- No study on the influence of TNF-α and Plasma Dobutamine concentration in patients Patients undergoing On-Pump CABG surgery.

  The results may help in risk stratification and may also guide inotrope dose adjustments in the intra and post operative period of On-Pump CABG surgery thereby reducing morbidity and mortality.

The objectives of this study are:

Primary Objective: Toevaluate if the preoperative level of the cardiac biomarker NT-Pro BNP, correlates with intraoperatively measured Pulmonary Capillary Wedge Pressure (PCWP) in patients undergoing On-Pump CABG surgery.

Secondary Objective: To evaluate the role of the inflammatory mediator TNF alpha on the postoperative plasma concentration of Dobutamine

Tertiary Objective : To evaluate the role of NT-pro BNP and TNF-α on existing comorbidities.

Sample size: 44 patients

Study participants: All patients undergoing On-Pump CABG surgery in CTVS Department of PSG Hospital**.**

Location: Department of CTVS , PSG Hospital, Coimbatore

We request you to kindly cooperate with us in this study. We propose to collect background information and other relevant details related to this study. We will be carrying out:

Initial interview: 10 to 15 minutes.

Data collected will be stored for a period of fifteen years. We will / will not use the data as part of another study.

Blood sample collection: 4 ml directly from patient or from left over samples in Pathology and Biochemistry labs, these collected samples will not be used for any other purposes

No. of times it will be collected: **Twice**

Whether blood sample collection is part of routine procedure or for research (study) purpose: **Research purpose**

Specify **purpose**, discomfort likely to be felt and side effects, if any: **To determine NT-Pro BNP, TNF-α and plasma concentration of dobutamine .No discomfort or side effects.**

Whether blood sample collected will be stored after study period**: No**
Case details and data will be stored for 5 yrs
Whether blood sample collected will be sold: **No**
Whether blood sample collected will be shared with persons from another institution:**No**

Medication given, if any, duration, side effects, purpose, benefits**: No medications**
Benefits from this study: Correlation of Preoperative NT-Pro BNP with PCWP would help in risk stratification and correlation of TNF-α with Plasma concentration of dobutamine may guide inotrope dose adjustments in the intra and post operative period of On-Pump CABG surgery thereby reducing morbidity and mortality.

Risks involved by participating in this study: **No risks**
How the **results** will be used: the results will be used for **further researches and publications**

If you are uncomfortable in answering any of our questions during the course of the interview / biological sample collection, **you have the right to withdraw from the interview / study at anytime.** You have the freedom to withdraw from the study at any point of time. Kindly be assured that your refusal to participate or withdrawal at any stage, if you so decide, will not result in any form of compromise or discrimination in the services offered nor would it attract any penalty. You will continue to have access to the regular services offered to a patient. You will **NOT** be paid any remuneration for the time you spend with us for this interview / study. The information provided by you will be kept in strict confidence. Under no circumstances shall we reveal the identity of the respondent or their families to anyone. The information that we collect shall be used for approved research purposes only. You will be informed about any significant new findings - including adverse events, if any, – whether directly related to you or to other participants of this study, developed during the course of this research which may relate to your willingness to continue participation.

Consent: The above information regarding the study, has been read by me/ read to me, and has been explained to me by the investigator/s. Having understood the same, I hereby give my consent to them to interview me. I am affixing my signature / left thumb impression to indicate my consent and willingness to participate in this study (i.e., willingly abide by the project requirements).

Signature / Left thumb impression of the Study Volunteer / Legal Representative:

Signature of the Interviewer with date: Witness:
Contact no: 8754272347

பூ. சா. கோ மருத்துவக் கல்லூரி மற்றும் ஆராய்ச்சி நிறுவனம், கோவை

மனித நெறிமுறைக் குழு

ஒப்புதல் படிவம்

தேதி:

மரு. இரா. சுபாஷினி, ஆகிய நான் பூ. சா. கோ மருத்துவக் கல்லூரியின் / மருத்துவமனையின் மருந்தியல் துறையின் கீழ், **"கோவையில் உள்ள ஒரு சூப்பர் ஸ்பெசாலிட்டி மருத்துவமனையில் ஓன்.பம்ப் சி.ஏ.பி.ஜி (பை பாஸ்) அறுவை சிகிச்சை செய்துக்கொள்ளும் நோயாளிகளின், உயிர் குறிப்பான்களுக்கும் (பயோமார்க்கர்ஸ்) நுரையீரல் கேப்பிலரி வெட்ஜ் அழுத்தம் மற்றும் பிளாஸ்மா டோபுடாமின் அளவுக்கும் உள்ள தொடர்பைக் கண்டறிதல்"** என்ற தலைப்பில் ஆய்வு மேற்கொள்ள உள்ளேன்.

என் ஆய்வு வழிகாட்டி: மரு. கே. புவனேஸ்வரி, மரு. பி.ஆர். முருகேசன்

ஆய்வு மேற்கொள்வதற்கான அடிப்படை:

ஓன்.பம்ப் சி.ஏ.பி.ஜி (பை பாஸ்) அறுவை சிகிச்சை செய்துக்கொள்ளும் நோயாளிகளின், உயிர் குறிப்பான்களுக்கும் (பயோமார்க்கர்ஸ்) நுரையீரல் கேப்பிலரி வெட்ஜ் அழுத்தம் மற்றும் பிளாஸ்மா டோபுடாமின் அளவுக்கும் உள்ள தொடர்பைக் கண்டறிதல்

ஆய்வின் நோக்கம்:

1. இதய உயிர்குறிப்பான் (பயோமார்க்கர்ஸ்) என்.டி.ப்ரோ பி.என்.பீக்கும் நுரையீரல் கேப்பிலரி வெட்ஜ் அழுத்தத்துக்கும் உள்ள தொடர்பு.
2. அழற்சி குறிப்பான் (டி.என்.ஃப்-ஆல்ஃபாவுக்கும் பிளாஸ்மா டோபுடாமின் அளவுக்கும் உள்ள தொடர்பு.
3. உடன் இருக்கும் மற்ற உடல்நல குறைகளுக்கும் மற்ற உயிர்குறிப்பான்களுக்கும் உள்ள தொடர்பு (சர்க்கரை நோய், ரத்த அழுத்தம், தைராய்டு)

ஆய்வில் பங்கு பெறும் நபர்களின் எண்ணிக்கை: 44

ஆய்வில் பங்கு பெறுவோர் மற்றும் வயது: ஓன்.பம்ப் சி.ஏ.பி.ஜி (பை பாஸ்) அறுவை சிகிச்சை செய்துக்கொள்ளும் நோயாளிகள் (40 வயதிலிருந்து 69வயது வரை).

ஆய்வு மேற்கொள்ளும் இடம்: பூ. சா. கோ. மருத்துவக்கல்லூரி மருத்துவமனை, கோயம்புத்தூர்.

இந்த ஆய்வில் எங்களுடன் ஒத்துழைக்குமாறு கேட்டுக்கொள்கிறோம். நாங்கள் சில தகவல்களை இந்த ஆய்விற்காக சேகரிக்க உள்ளோம்.

ஆய்வு செய்யப்படும் முறை:

முதன்மை நேர்காணல்: 10-15 நிமிடங்கள்

இந்த ஆய்வில் கிடைக்கும் தகவல்கள் **5 வருடங்கள்** பாதுகாக்கப்படும். இந்த தகவல்கள் வேறு ஆய்விற்குப் பயன்படுத்தப் பட மாட்டாது.

மருத்துவ பரிசோதனைகள்:

இரத்த மாதிரி சேகரிப்பு: 4மில்லி, இருமுறை

இரத்த மாதிரி எடுப்பது வழக்கமான சிகிச்சைக்காகவோ அல்லது இந்த ஆய்விற்காகவோ: குறிப்பிட்ட ஆய்விற்கக

இதனால் ஏற்படக் கூடிய அசௌகரியங்கள் / பக்க விளைவுகள்: இதனால் எந்த அசௌகரியமோ, பக்க விளைவுகளோ ஏற்படாது. **பொருந்தாது**

இரத்த மாதிரிகள் ஆய்விற்குப் பின் பாதுகாத்து வைக்கப்படுமா? ஆம் / இல்லை, அழிக்கப்படும்: **இல்லை**

சேகரிக்கப்பட்ட இரத்தம் விற்கப்படுமா? ஆம் / இல்லை **இல்லை**

சேகரிக்கப்பட்ட இரத்தம் வேறு நிறுவனத்துடன் பகிர்ந்து கொள்ளப்படுமா? ஆம் / இல்லை: **இல்லை**

மருந்துகள் ஏதேனும் கொடுக்கப்படவிருந்தால் அவை பற்றிய விவரம் (கொடுக்கப்படும் காரணம், காலம், பக்க விளைவுகள், பயன்கள்): **பொருந்தாது**

மருந்துகள் கொடுக்கப்படுவது வழக்கமான சிகிச்சை முறையா?: ஆம் / இல்லை (இல்லை என்றால் கொடுக்கப்படும் காரணம்) **பொருந்தாது**

கொடுக்கப்படும் மருந்துகளுக்கு மாற்று உள்ளதா?: ஆம் / இல்லை (ஆம் என்றால் இந்த குறிப்பிட்ட மருந்து கொடுக்கப்படும் காரணம்) **பொருந்தாது**

ஆய்வில் பங்குபெறுவதால் ஏற்படும் பலன்கள்:

இந்த ஆய்வின் முடிவில் என்.டி.ப்ரோ பி.ன்.பீ மற்றும் நுரையீரல் கேப்பிலரி வெட்ஜ் அழுத்தத்திற்கும் உள்ள தொடர்பை கொண்டு ஒன் பம்ப் சி.ஏ.பி.ஜி அறுவை சிகிச்சை செய்து கொள்ளும் நோயாளிகளை ஆபத்து வகைப்படுத்த உதவும். டோபுடாமின் மற்றும் டி.என்.ஃப்-ஆல்ஃபா விற்கும் உள்ள தொடர்பை வைத்து அறுவை சிகிச்சையின் போது டோபுடாமின் அளவை சரி செய்ய உதவும்.

ஆய்வில் பங்கேற்பதால் ஏற்படும் அசௌகரியங்கள் / பக்க விளைவுகள்: இந்த ஆய்வினால் தங்களுக்கு எந்த விதமான அபாயங்களும் அசௌகரியங்களும் ஏற்படாது.

ஆய்வின் முடிவுகள் எந்த முறையில் பயன்படுத்தப்படும்?

ஆய்வின் முடிவுகள், அடுத்தகட்ட ஆராய்ச்சிகளுக்கும், மருத்துவ ஆய்வு பத்திரிக்கைகளில் வெளியிடுவதற்கும் பயன்படுத்தப்படும்..

இந்த ஆய்வின் கேள்விகளுக்கு பதிலளிப்பதோ, இரத்த மாதிரிகள் அல்லது திசு மாதிரிகள் எடுப்பதிலோ உங்களுக்கு ஏதேனும் அசௌகரியங்கள் இருந்தால், எந்த நேரத்தில் வேண்டுமானாலும் ஆய்விலிருந்து விலகிக்கொள்ளும் உரிமை உங்களுக்கு உண்டு. ஆய்விலிருந்து விலகிக்கொள்வதால் உங்களுக்கு அளிக்கப்படும் சிகிச்சை முறையில் எந்த வித பாதிப்பும் இருக்காது என்று உங்களுக்கு உறுதியளிக்கிறோம். மருத்துவ மனையில் நோயாளிகளுக்கு அளிக்கப்படும் சேவைகளை நீங்கள் தொடர்ந்து பெறலாம். இந்த ஆய்வில் பங்கேற்க ஒப்புக்கொள்ளுவதால் வேறு எந்த விதமான கூடுதலான பலனும் உங்களுக்குக் கிடைக்காது. நீங்கள் அளிக்கும் தகவல்கள் இரகசியமாக வைக்கப்படும். ஆய்வில் பங்கேற்பவர்கள் பற்றியோ அவர்கள் குடும்பத்தைப் பற்றியோ எந்தத் தகவலும் எக்காரணம் கொண்டும் வெளியிடப்படாது என்று உறுதியளிக்கிறோம். நீங்கள் அளிக்கும் தகவல்கள் / இரத்த மாதிரிகள் / திசு மாதிரிகள் அங்கீகரிக்கப்பட்ட ஆய்விற்கு மட்டுமே பயன்படுத்தப்படும். இந்த ஆய்வு நடைபெறும் காலத்தில் குறிப்பிடத்தகுந்த புதிய கண்டுபிடிப்புகள் அல்லது பக்க விளைவுகள் ஏதும் ஏற்பட்டால் உங்களுக்குத் தெரிவிக்கப்படும். இதனால் ஆய்வில் தொடர்ந்து பங்கு பெறுவது பற்றிய உங்கள் நிலைப்பாட்டை நீங்கள் தெரிவிக்க ஏதுவாகும்.

ஆய்வுக்குட்படுபவரின் ஒப்புதல்: இந்த ஆய்வைப் பற்றிய மேற்கூறிய தகவல்களை நான் படித்து அறிந்து கொண்டேன் / ஆய்வாளர் படிக்கக் கேட்டுத் தெரிந்து கொண்டேன். ஆய்வினைப் பற்றி நன்றாகப் புரிந்து கொண்டு இந்த ஆய்வில் பங்கு பெற ஒப்புக்கொள்கிறேன். இந்த ஆய்வில் பங்கேற்பதற்கான எனது ஒப்புதலை கீழே கையொப்பமிட்டு, கை ரேகை பதித்து நான் தெரிவித்துக் கொள்கிறேன்.

பங்கேற்பாளரின் பெயர், முகவரி:

பங்கேற்பாளரின் கையொப்பம் / கை ரேகை / சட்டப்பூர்வ பிரதிநிதியின் கையொப்பம்:

தேதி :

ஆய்வாளரின் கையொப்பம்:

தேதி :

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A Study To Evaluate The Association Of Biomarkers With Pulmonary Capillary Wedge Pressure And Plasma Concentration Of The Inotrope Dobutamine In Patients Undergoing On-Pump CABG Surgery In A Super-Speciality Hospital At Coimbatore : A Prospective Observational Study

Dr.R.Subhashini, Dr. K. Bhuvaneswari, Dr.P.R.Murugesan

Case Proforma

Patient name: IP/OP no:

Age/Gender: Address:

Contact number:

Height: Weight: BMI:

Occupation (Current &Past):

Ethnicity (Religion/caste) :

History of Presenting Illness :

Past History:

H/o Hypertension, Diabetes mellitus, Coronary Artery Disease, Congestive Cardiac Failure,Thyroid disorder ,Liver and Renal disorders

H/o Surgery

Drug History:

H/o chronic anti-inflammatory drug intake, Recombinant BNP analogues

Personal History:

Smoking/Alcohol -

Other Substance abuse-

Family History:

H/O Cardiovascular diseases / CABG

Menstrual History:

LMP:

(Including age at menarche, menopause attained/not)

DIAGNOSIS :

Pre Operative Investigations	**CABG Surgery details**	**Intraoperative Parameters Assessed**	**Post-Operative Investigations**

DATA COLLECTION TOOL

PRIMARY OBJECTIVE:

PRE–OPERATIVE NT PRO BNP VALUE (ng/ml)	INTRAOPERATIVE PCWP (mm Hg)	OTHER INTRAOPERATIVE PARAMETERS
		• Cardiopulmonary Bypass Time: (mins) • Cardioplegia: Anterograde/Retrograde • Pulse : Average (bpm) • BP: Average (mm Hg) • CVP: Average (mm Hg) • IABP : Needed / Not needed

SECONDARY OBJECTIVE:

PRE–OPERATIVE	INTRAOPERATIVE (Sample collected 4th hr from Skin Incision)		
TNF α VALUE (ng/ml)	Dobutamine Dose Administered by infusion pump (ng/ml)	TNF –α (ng/ml)	Dobutamine concentration in Plasma– Measured using HPLC (ng/ml)