Final submission of Research Report

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Student number: 980 3048x

20th September 2013

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Mmed NUCLEAR MEDICINE

EVALUATION OF THE EFFICACY OF FULL FAT MILK AND DILUTED LEMON JUICE VERSUS NO INTERVENTION TO REDUCE INTERFERING INFRA-CARDIAC ACTIVITY OF TC-99M SESTAMIBI DURING MYOCARDIAL PERFUSION IMAGING

LIST OF CORRECTIONS

1. Cover page: Date changed to September 2013

2. Page iii
   P3: Mmed Nuclear Medicine
   P4: This letter is to certify that Dr. K. Purbhoo has done her research in Nuclear Medicine.

3. Page vii
   P1: The use of Single Photon Emission Computed Tomography (SPECT) myocardial perfusion imaging (MPI), with Technetium – 99m (Tc-99m) Sestamibi in conjunction with either exercise, pharmacologic stress or both is an established tool for both the diagnosis and prognostication of patients with ischemic heart disease.
   P1: For perfusion imaging with SPECT, Tc-99m labeled radiopharmaceuticals (Sestamibi or Tetrofosmin) are commonly used.
   P3: A total of six hundred and thirty (630) patients who fulfilled the inclusion criteria were randomized without stratification into three groups. Group 0 (G0) were given diluted lemon juice, 246 patients; full fat milk to group 1 (G1), 313 patients and group 2 (G2); 71 patients, had no intervention. The latter was the control group. Raw data of both the stress and rest images were visually and quantitatively assessed by two Nuclear Medicine physicians for the presence of infra-cardiac activity.

4. Page ix
   Table of Contents: added Nomenclature and Abbreviations
   Amended point 2.3: Patient preparation and Stress Protocol
   Amended point 2.4: Imaging Protocol and data processing

5. Page xiii
   SPECT/CT- Single Photon Emission Computed Tomography with Computed Tomography
   FBP – Filtered backprojection
   CCK - Cholecystokinin
   KeV – Kiloelectron Volt

MBq – Megabequerel
mSv - Millisievert
CVD – Cardiovascular Disease
LAO – Left Anterior Oblique
LV – Left Ventricle
ECG – Electrocardiogram

6. Page 1
   P2: The use of Single Photon Emission Computed Tomography (SPECT) myocardial perfusion imaging (MPI), with Technetium (Tc-99m) Sestamibi in conjunction with either exercise, pharmacologic stress or both, is an established tool for both the diagnosis and prognostication of patients with ischemic heart disease.[1]
   P3: For perfusion imaging with SPECT, Thallium-201 (Tl-201) andTechnetium-99m (Tc-99m) labeled radiopharmaceuticals (Sestamibi or Tetrofosmin) are commonly used.
   P3: Technetium -99m labeled compounds do not have these limitations, which have encouraged their development and increasing use of them.
   P4: About 27% of the injected activity is cleared through renal elimination after 24 hours and 33% of the injected activity is cleared through the fecal route in 48 hours

7. Page 2
   P1: This may falsely elevate the number of counts the camera assigns to the nearby cardiac wall, thus displaying the cardiac region as falsely 'hotter'. Both artifacts are patient dependent and their severity is hard to predict.
   P2: When using Tc-99m-Sestamibi cardiac SPECT imaging in the evaluation of myocardial perfusion, the presence of infra-cardiac activity leads to artifacts, reducing the desired target to background ratio.
   P3: Several different protocols, including eating a fatty meal; drinking milk, milk and water, milkshakes, or iodinated oral contrast; intravenous injection of cholecystokinin (CCK), and the administration of metoclopramide and erythromycin have been studied in an attempt to reduce the artifacts arising from abdominal activity. These substances are thought to stimulate gallbladder contraction and therefore liver clearance, and should improve visualization of the inferior myocardial wall.

8. Page 3
   P1: Sestamibi requires constitution with Tc-99m-sodium pertechnetate which is produced from a Molybdenum generator.
   P1: Sestamibi is lipophilic and diffuses passively through capillary and cell membranes. Once within the myocardium it is localized mostly in mitochondria where it is trapped. Retention is based on intact mitochondria, thus reflecting viable myocytes.
   P2: The physical half- life of Tc-99m requires that imaging be completed by 6 hours. After a resting intravenous injection, the highest extra-cardiac activity is seen in the gallbladder and liver, followed by the kidneys. Myocardial uptake, which is coronary flow dependent, is 1.5% of the injected dose at stress and 1.2% of the injected dose at rest.

P3: Dynamic exercise is the technique of choice for the assessment of patients with suspected or known coronary artery disease, provided that the patient is able to exercise to an acceptable work load of 85% expected heart rate for age.

9. Page 4
   P2: Two groups of drugs commonly used as substitutes for exercise stress testing include: vasodilators (dipyridamole and adenosine) that create coronary hyperemia, and the sympathomimetic agents (dobutamine and arbutamine) which increase myocardial oxygen demand.
   P3: Dipyridamole induces coronary and peripheral vasodilatation.
   P3: Myocardium supplied by a diseased coronary artery has reduced perfusion reserve and this leads to heterogeneity of perfusion during vasodilation or even to myocardial ischemia caused by "coronary steal".
   P3: As a routine, if patients are able to walk and they have no contraindications to exercise, they are exercised for about 4 minutes after pharmacological infusion with vasodilators.
   P3: Patients are required to abstain from caffeine- containing products for 48 hours prior to MPI in case a vasodilator stress needs to be conducted.

10. Page 5
    P1: It is a secondary pharmacological stressor that is used in patients who cannot undergo exercise stress and have contraindications to vasodilator agents. During Dobutamine intravenous infusion, the presence of a coronary stenosis limits the flow increase that can occur through that vessel, leading to flow heterogeneity and/or the appearance of a left ventricular wall motion abnormality (resulting from imbalance between myocardial oxygen supply and demand).
    P3: Whole milk is believed to stimulate liver clearance as well as increasing peristaltic movement.

11. Page 6
    P2: The proximity of the heart to these organs contributes to the interference which complicates the interpretation of the inferior and infero-septal walls of the left ventricle.

12. Page 8
    P1: Late imaging does not have any significant effect on the functional parameters of the MPI, including the ejection fraction, however due to the half- life of Tc-99m (6h), studies need to be completed before significant decay of the radiotracer may compromise image quality and influence clinical interpretation.

13. Page 9
    Table 1: Exclusion Criteria:
    - Inability to drink 250 ml of fluids secondary to medically essential fluid restriction
    - Pregnancy
    - Previous cholecystectomy and/or liver or biliary system disease

    Point 2.3: Patient preparation and stress protocol

P2: Patients were fasted at least four hours prior to stress testing (usually overnight) and were required to abstain from caffeine- containing beverages and methylxanthine containing medications for at least 24 hours. The reason is that caffeine and methylxanthines block the adenosine receptors on arterial smooth muscle cells, thus limiting the effectiveness of vasodilator agents.

14. Page 10
    P2: Generally, routine imaging for stress is carried out 30 – 45 minutes post tracer injection, however in this study some patients were imaged later due to the longer acquisition times with the addition of prone imaging, which is a routine protocol in our department.
    Point 2.4: Imaging protocol and Data Processing

15. Page 11
    P1: Two experienced Nuclear Medicine physicians evaluated the raw data of the anterior (ANT) and left lateral (LLAT) views of both the stress and rest studies of all study participants for the presence or absence of interfering infra-cardiac activity.
    P2: Visual and semi-quantitative assessment of the raw data of both stress and rest images as previously used by Hofman *et al* was used.

16. Page 16
    Table 4: Grading: 0: absent infra-cardiac activity; 1: infra-cardiac activity < myocardial activity; 2: infra-cardiac activity = myocardial activity; 3: infra-cardiac activity > myocardial activity.

17. Page 18
    P2: As mentioned earlier the use of Tc-99m Sestamibi for MPI often results in the increased splanchnic activity that may create a major problem in the visual and quantitative interpretation of inferior and infero-septal walls of the left ventricle.

18. Page 21
    P1: Our study demonstrated that the administration of milk or lemon juice results in a significant decrease in the intensity of infra-cardiac activity as compared to the group with no intervention. At stress, both the milk group and the lemon juice group were comparable. At rest, the milk group showed a trend toward greater improvement than the group that received lemon juice. However, we are not sure on the impact of the amount of either milk or lemon juice as well as their timing of administration. Therefore, further studies are needed to determine the ideal amount and timing for administration of these interventions.

19. Page 27
    P5: A minimum of 500 patients will be enrolled in the study.

20. Page 29
    P1: Objectives: When using Tc-99m Sestamibi for myocardial perfusion imaging, the increased infra-cardiac activity may create a major problem in the visual and quantitative interpretation of the inferior and infero-septal walls of the left ventricle.

21. Page 32
    Heading number 2 amended: Patient preparation and Stress protocol

P2: Patients were fasted at least four hours prior to stress testing (usually overnight) and were required to abstain from caffeine- containing beverages and methylxanthine containing medications for at least 24 hours. The reason is that caffeine and methylxanthines block the adenosine receptors on arterial smooth muscle cells, thus limiting the effectiveness of vasodilator agents.
P3: Generally routine imaging for stress is carried out 30 – 45 minutes post tracer injection, however in this study some patients were imaged later due to the longer acquisition times with the addition of prone imaging, which is also a routine protocol in our department.

22. Page 33
    Heading amended: Imaging protocol and Data Processing
    P3: Two experienced Nuclear medicine physicians evaluated the raw data of the anterior (Ant) and left lateral (LLAT) views of both the stress and rest studies of all study participants for the presence or absence of interfering infra-cardiac activity.

23. Page 36
    P5: As mentioned earlier the use of Tc-99m Sestamibi for MPI often results in increased splanchnic activity that may create a major problem in the visual and quantitative interpretation of inferior and infero-septal walls of the left ventricle.

24. Page 44
    Table 4: Grading: 0: absent infra-cardiac activity; 1: Infra-cardiac activity < myocardial activity; 2: Infra-cardiac activity = myocardial activity; 3: Infra-cardiac activity > myocardial activity.

EVALUATION OF THE EFFICACY OF FULL FAT MILK AND DILUTED LEMON JUICE VERSUS NO INTERVENTION TO REDUCE INTERFERING INFRA-CARDIAC ACTIVITY OF TC-99M SESTAMIBI DURING MYOCARDIAL PERFUSION IMAGING

Dr. Khushica Purbhoo

A Research Report submitted to the Faculty of Health Sciences, University of the Witwatersrand, in fulfillment of the requirements for the degree of

Master of Medicine

In the branch of Nuclear Medicine

Johannesburg

September 2013

Candidate's declaration

I, Khushica Purbhoo declare that this research report is my own work. This research report is being submitted for the degree of Master of Medicine in the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination at this or any other University.

20th September 2013

Master of Medicine in the branch of Nuclear Medicine

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28 February 2013

To whom it may concern

Re: Dr. Khushica Purbhoo
Student number: 9803048x
Staff number: A0004224
Mmed Nuclear medicine

This letter is to certify that Dr. K. Purbhoo has done her research in Nuclear medicine. Her topic is **"Evaluation of the efficacy of full fat milk and diluted lemon juice versus no intervention to reduce interfering infra-cardiac activity of Tc-99m Sestamibi during myocardial perfusion imaging."** She recruited patients to her study that were referred for Myocardial Perfusion Imaging and followed her protocol for her study accordingly.
Her data was entered into an excel sheet and she has solely written the work presented on her own. The entire research article was done by herself with assistance from her supervisor.

Kind Regards,

Prof MDTHW Vangu
Head of Department
Nuclear Medicine

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Dedication

- **My parents, Pramod and Sooshila Purbhoo:** Thank you for your unconditional support with my studies. I am honoured to have you as my parents. Thank you for giving me the chance to improve myself through all my walks of life. I love you.

- **My husband, Regan and my sons, Eashaan and Trishul:** Thank you for believing in me; for allowing me to further my career opportunities. Please do not doubt my dedication and unconditional love for you.

Acknowledgements

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- A special thanks to Professor Elena Libhaber for her assistance with the statistics.
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- The work was supported in part by a grant received from the University of the Witwatersrand, South Africa.

Paper for publication: to be submitted from this thesis

Evaluation of the efficacy of full fat milk and diluted lemon juice versus no intervention to reduce interfering infra-cardiac activity of Tc-99m Sestamibi during Myocardial Perfusion Imaging

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Abstract

The use of Single Photon Emission Computed Tomography (SPECT) myocardial perfusion imaging (MPI), with Technetium – 99m (Tc-99m) Sestamibi in conjunction with either exercise, pharmacologic stress or both is an established tool for both the diagnosis and prognostication of patients with ischemic heart disease. For perfusion imaging with SPECT, Tc-99m labeled radiopharmaceuticals (Sestamibi or Tetrofosmin) are commonly used. The major metabolic pathway for clearance of Sestamibi is the hepatobiliary system which creates difficulty in both visual and quantitative interpretation of myocardial perfusion, particularly of the inferior and infero-septal walls after reconstruction.

Diluted lemon juice, an acid-rich drink is an alimentary cholekinetic that facilitates Sestamibi transit through the liver. Whole milk stimulates liver clearance as well as increases peristaltic movement. The aim of the study was to determine which protocol would be the best to reduce interfering infra-cardiac activity and therefore result in an improvement in image quality. We had three groups, comparing the use of full fat milk, diluted lemon juice and a control group that had no intervention.

All patients referred to our institution for MPI from November 2009 to May 2012 were enrolled in the study. A total of six hundred and thirty (630) patients who fulfilled the inclusion criteria were randomized without stratification into three groups. Group 0 (G0) were given diluted lemon juice, 246 patients; full fat milk to group 1 (G1), 313 patients and group 2 (G2); 71 patients, had no intervention. The latter was the control group. Raw data of both the stress and rest images were visually and quantitatively assessed by two Nuclear Medicine physicians for the presence of infra-cardiac activity. The physicians were blinded to the intervention received and the data were reviewed simultaneously.

The administration of milk or lemon juice resulted in a significant decrease in the intensity of infra-cardiac activity compared to the control group. This improvement was even more significant in the milk group for patients done during rest myocardial perfusion imaging

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Nomenclature and Abbreviations

MIBI – Methoxy-isobutyl Isonitrile (Sestamibi)

MPI – Myocardial Perfusion Imaging

SPECT – Single Photon Emission Computed Tomography

SPECT/CT- Single Photon Emission Computed Tomography with Computed Tomography

Tc-99m – Technetium 99 metastable

Tl-201 – Thallium 201

FBP – Filtered backprojection

CCK - Cholecystokinin

KeV – Kiloelectron Volt

MBq – Megabequerel

mSv - Millisievert

CVD – Cardiovascular Disease

LAO – Left Anterior Oblique

LV – Left Ventricle

ECG – Electrocardiogram

1.0 INTRODUCTION

Coronary artery disease is a major cause of death throughout the world.

The use of Single Photon Emission Computed Tomography (SPECT) myocardial perfusion imaging (MPI), with Technetium (Tc-99m) Sestamibi in conjunction with either exercise, pharmacologic stress or both, is an established tool for both the diagnosis and prognostication of patients with ischemic heart disease.[1] Patients with normal perfusion on Tc-99m SPECT MPI have an excellent prognosis, whereas patients with abnormal perfusion have an increased rate of cardiac death and non-fatal infarction during follow-up.[2] This approach is based on the physiologic alterations of regional myocardial blood flow and contraction caused by exercise or pharmacologic stress. They are more predictive of outcome than the coronary anatomy alone.[3] A non-invasive approach is safer and less expensive than an invasive investigation such as cardiac catherization with coronary angiography.[3]

For perfusion imaging with SPECT, Thallium-201 (Tl-201) and Technetium-99m (Tc-99m) labeled radiopharmaceuticals (Sestamibi or Tetrofosmin) are commonly used. Tl-201 has many limitations compared to Tc-99m. It has a long physical half- life(73 hours) and delivers a high radiation burden to the patient(80MBq delivers an effective dose of 18mSv, higher than coronary angiography).[4] The low injected activity results in a low target to background ratio, therefore images can be sub-optimal and impair high quality ECG gated SPECT studies. The low energy emission (135keV, 167keV with 12% abundance), results in significant attenuation by soft tissue.[4] Technetium -99m labeled compounds do not have these limitations, which have encouraged their development and increasing use of them.

The major metabolic pathway for clearance of Tc-99m Sestamibi is the hepatobiliary system. Activity from the gallbladder appears in the intestine within one hour of injection. About 27% of the injected activity is cleared through renal elimination after 24 hours and 33% of the injected activity is cleared through the fecal route in 48 hours.[4]

Filtered backprojection (FBP) is the technique used to process the MPI data. With FBP, image artifacts can be caused by halo and spillover effects. The halo effect is a reconstruction artifact due to the implementation of FBP on attenuation projection images, with focally increased activity in low count surroundings. This results in an underestimation of the inferior and infero-septal walls. Spillover of activity into the myocardium, resulting from photon scatter is associated with activity in the liver, bowel and stomach and may result in an overestimation of the counts in the inferior wall of the left ventricle. This may falsely elevate the number of counts the camera assigns to the nearby cardiac wall, thus displaying the cardiac region as falsely 'hotter'. Both artifacts are patient dependent and their severity is hard to predict.[5]

When using Tc-99m-Sestamibi cardiac SPECT imaging in the evaluation of myocardial perfusion, the presence of infra-cardiac activity leads to artifacts, reducing the desired target to background ratio. This creates difficulty in both visual and quantitative interpretation of myocardial perfusion particularly of the inferior and infero-septal walls.[1] Infra-cardiac activity is less common with exercise and more common with pharmacological stress and rest images.[2]

Several different protocols, including eating a fatty meal; drinking milk, milk and water, milkshakes, or iodinated oral contrast; intravenous injection of cholecystokinin (CCK), and the administration of metoclopramide and erythromycin have been studied in an attempt to reduce the artifacts arising from abdominal activity. These substances are thought to stimulate gallbladder contraction and therefore liver clearance, and should improve visualization of the inferior myocardial wall. The conclusive benefits, however, are controversial.[1,3,5,6,7,8,9,10,11,12,13]

1.1 Background

Tc-99m Sestamibi is a monovalent cation which consists of six methoxyisobutyl-isonitrile (MIBI) ligands surrounding a technetium central core.[4] Sestamibi requires constitution with Tc-99m-sodium pertechnetate which is produced from a Molybdenum generator. Tc-99m decays by isomeric transition to Tc-99m at a lower energy with a half- life of 6.03h, emitting mostly (89%) gamma photons of 140keV.[4] Sestamibi is lipophilic and diffuses passively through capillary and cell membranes. Once within the myocardium it is localized mostly in mitochondria where it is trapped. Retention is based on intact mitochondria, thus reflecting viable myocytes. It undergoes minimal redistribution (10-15%) from its initial pattern of uptake.[4]

Following physical or pharmacological stress, the highest initial concentration of the administered Sestamibi in descending order is seen in the gallbladder, heart, liver and spleen. Gallbladder activity peaks at 60 minutes and then gradually decreases.[4] Other extra-cardiac activity progressively reduces from the time of injection. Cardiac activity remains relatively stable. The physical half- life of Tc-99m requires that imaging be completed by 6 hours. After a resting intravenous injection, the highest extra-cardiac activity is seen in the gallbladder and liver, followed by the kidneys.[1,4] Myocardial uptake, which is coronary flow dependent, is 1.5% of the injected dose at stress and 1.2% of the injected dose at rest.[1,4] The post-stress SPECT imaging usually begins 30-60 minutes after tracer injection to allow for hepatobiliary clearance, with a longer delay required for resting images and for pharmacological stress with vasodilators because of higher liver uptake.[4]

Stress Myocardial Perfusion Imaging

Dynamic exercise is the technique of choice for the assessment of patients with suspected or known coronary artery disease, provided that the patient is able to exercise to an acceptable work load of 85% expected heart rate for age. It also gives information about exercise tolerance.[1,2,14] Using the treadmill,

Bruce protocol, the speed and gradient increases every 3 minutes to provide an intense workload over a relatively short period of time. For patients with lower exercise capacity a modified Bruce protocol is an alternative.[2] Complications as a result of exercise testing are rare, (approximately 1:10,000).[1]

About 40 – 50% of patients referred for stress testing are unable to perform adequate physical exercise on a motor driven treadmill.[2] These are patients with neurologic, orthopedic, respiratory or vascular problems. A standard exercise test has been shown to be a suboptimal means for their assessment leading to reduced sensitivity.[2] This problem has led to the development of pharmacological stress agents, which remove the need for patient co-operation and enable a confident assessment of cardiac function. Two groups of drugs commonly used as substitutes for exercise stress testing include: vasodilators (dipyridamole and adenosine) that create coronary hyperemia, and the sympathomimetic agents (dobutamine and arbutamine) which increase myocardial oxygen demand.[2]

Dipyridamole induces coronary and peripheral vasodilatation. The increase in coronary flow at a dose of 140µg/kg/min is 4.4 times baseline.[2] Dipyridamole is an indirect coronary arteriolar dilator that increases the tissue levels of adenosine. Myocardium supplied by a diseased coronary artery has reduced perfusion reserve and this leads to heterogeneity of perfusion during vasodilation or even to myocardial ischemia caused by "coronary steal".[2] One of the limitations of vasodilators is the high splanchnic uptake, which may create artifactual myocardial perfusion defects after filtered backprojection. As a routine, if patients are able to walk and they have no contraindications to exercise, they are exercised for about 4 minutes after pharmacological infusion with vasodilators. This helps improve the side effects of the vasodilator and improves bowel clearance.[2] Patients are required to abstain from caffeine- containing products for 48 hours prior to MPI in case a vasodilator stress needs to be conducted.

Dobutamine is a synthetic sympathomimetic amine that stimulates ß-adrenergic receptors and produces an increase in myocardial contractility and a secondary increase in myocardial blood flow.[2] It is a secondary pharmacological stressor that is used in patients who cannot undergo exercise stress and have contraindications to vasodilator agents. During Dobutamine intravenous infusion, the presence of a coronary stenosis limits the flow increase that can occur through that vessel, leading to flow heterogeneity and/or the appearance of a left ventricular wall motion abnormality (resulting from imbalance between myocardial oxygen supply and demand).[2]

In this study, both dipyridamole and dobutamine were used as required by patient referral.

1.2 Study Objectives

Physiologically, acid-rich food or drink has the potential to facilitate hepatobiliary clearance of bile by increasing the secretion of secretin.[9] Diluted lemon juice is an acid-rich drink that is used as an alimentary cholekinetic stimulus to stimulate Sestamibi transit through the liver. Whole milk is believed to stimulate liver clearance as well as increasing peristaltic movement.[8] A comparison was made between the diluted lemon juice and full fat milk compared to a group with no intervention. The aim of this study was to determine:

- What percentage of patients referred for MPI had interfering infra-cardiac activity at rest and post stress?
- The influence of drinking lemon juice or full fat milk on Tc-99m Sestamibi transit through the liver
- Which protocol, if any, between milk and lemon juice would be better for reducing infra-cardiac activity

1.3 Literature Review

Myocardial Perfusion Imaging (MPI) is considered an important non-invasive diagnostic and prognostic tool in the evaluation of ischemic heart disease. It is capable of identifying abnormalities in perfusion and provides information on myocardial function.

Myocardial perfusion agents labeled with Tc-99m, such as Sestamibi are clinically validated for MPI.[4] The tracer is mainly excreted by hepatobiliary clearance and this upper abdominal activity adjacent to the myocardium may cause artifacts in the interpretation of the perfusion images. The heart lies on the diaphragm, just above the left lobe of the liver and in the vicinity of the bowel. The proximity of the heart to these organs contributes to the interference which complicates the interpretation of the inferior and infero-septal walls of the left ventricle.It can result in increased or decreased tracer uptake in these areas in the visual and quantitative assessment after reconstruction with FBP in SPECT studies.[1,2,4] The interfering activity also results in interference with the quantification of wall motion which can result in large areas of apparently decreased activity due to inappropriate count normalization and increased image variability. Extra-cardiac activity is also influenced by the type of stress. Stress with exercise results in significantly reduced liver activity when compared with rest studies or stress with pharmacological methods.[2,4]

A number of studies have explored the opportunities to reduce upper abdominal activity in MPI.[5,6,7,8,9,10,11,12,13] Most of the research has focused on a role of different liquids and solid food, especially fatty meals, positioning, as well as the time from injection to image acquisition, to assist in hepatobiliary clearance. The proposed impetus by using these methods is (1) to fill the stomach thereby increasing the distance between the left ventricle and interfering infra-cardiac activity, (2) increase liver

clearance of tracer via gallbladder contraction. The results have shown significant impact of individual variables but less information is available about a combination of more than one variable. Studies have also examined the degree of intestinal activity using quantitative measures such as lung/myocardial or liver/myocardial ratios.[9,10]

With the routine use of Tc-99m Sestamibi in our department, it was found that a large number of patients had interfering bowel activity and the study was either repeated or prone images, which are also routinely done, helped in the interpretation of inferior/infero-septal wall artifacts. Studies have proven that acquisition in the prone position has shown an improvement in the specificity of visually analyzed myocardial perfusion SPECT. Patients with inferior wall defects on supine MPI that are not present on prone MPI have a lower risk of subsequent cardiac events. [7]

In a study by Lyngholm *et al*[15], comparing the interaction of food and time on intestinal activity, it was found that gastric filling and delayed image acquisition reduced upper abdominal activity by utilizing the volume effect and therefore reduced the need for repeat scans. With the implementation of this intervention a success rate of 98.4% (127 of 129 patients) at stress and 98.8% (84 of 85 patients) at rest was achieved. A study by Hofman *et al*[6], comparing milk versus water in reducing infra-cardiac activity showed that milk rather than water resulted in a significant decrease in intensity of infra-cardiac activity. However the reduction in the intensity of infra-cardiac activity in the milk or water group did not translate into a statistically significant benefit in the image ($p= 0.563$ at stress and $p= 0.502$ at rest).

It has been documented that Sestamibi is cleared from the liver at a greater rate than from the myocardium. Thus, a longer delay from radiopharmaceutical injection to image acquisition will result in

reduced interference from liver activity.[4,8] Late imaging does not have any significant effect on the functional parameters of the MPI, including the ejection fraction, however due to the half- life of Tc-99m (6h), studies need to be completed before significant decay of the radiotracer may compromise image quality and influence clinical interpretation.

Overall, conclusive results comparing different techniques used to increase clearance of infra-cardiac activity have been controversial and therefore we chose to look at the effects of diluted lemon juice or full fat milk compared to no intervention to assess the clearance of interfering infra-cardiac activity.

2.0 MATERIALS AND METHODS

Ethics approval was obtained from the University of Witwatersrand's Human Research Ethics Committee (HREC), ethics clearance number M091012.

2.1 Study population

All patients 18 years and older who were referred for MPI were enrolled in the study. Written consent was obtained from all study participants. The study commenced in November 2009 until May 2012. We recruited 904 patients in this study but data from 274 patients were excluded due to various reasons [non-return for second day study, milk or lemon juice not followed in a patient for both the stress and rest study and patients who fitted the exclusion criteria (Table 1)]. A total of 630 patients (304 female [48%] and 326 male [52%]), aged 19 – 84 years were eventually enrolled for data analysis.

Table 1. Inclusion and exclusion criteria

Inclusion Criteria	Exclusion Criteria
• Patients older than 18 years of age • Patients referred for Tc-99m Sestamibi Myocardial Perfusion scans	• Lactose intolerance • Patients who failed exercise and had a contraindication to pharmacologic stress testing ie. Vasodilators and dobutamine • Inability to drink 250 ml of fluids secondary to medically essential fluid restriction • Pregnancy • Previous cholecystectomy and/or liver or biliary system disease • Peptic ulcer disease within the last 6 months • History of diabetes mellitus • Previous myocardial infarction within the last 2 months, unstable angina, severe primary valvular disease, left ventricular aneurysm, primary cardiomegaly, left ventricle hypertrophy or severe conduction disturbances

2.2 Randomization

Patients were randomized into three groups. Group 0 (G0) had diluted lemon juice, group 1 (G1) had full fat milk and group 2 (G2) had no intervention (control group). Full fat milk consisted of 250mL milk. Diluted lemon juice consisted of 50mL lemon juice and 200mL water, total volume of 250mL.

2.3 Patient preparation and stress protocol

A routine two day protocol was used. Patients were stressed on day one and a rest study was done on day two. Patients were fasted at least four hours prior to stress testing (usually overnight) and were required to abstain from caffeine- containing beverages and methylxanthine containing medications for at least 24 hours. The reason is that caffeine and methylxanthines block the adenosine receptors on arterial smooth muscle cells, thus limiting the effectiveness of vasodilator agents. Beta-blockers and

calcium channel antagonists were also withheld. Our department protocol is that we withhold caffeine in all patients even if exercise stress is planned in case there is a necessity to switch to pharmacological stress. The patients were hemodynamically and clinically stable for 48 hours prior to the test.

The stress modality (Treadmill, Dipyridamole or Dobutamine) was chosen and implemented in accordance with the recent EANM guideline.[14] Following the injection of 740MBq Tc-99m Sestamibi during stress, patients in G0 received diluted lemon juice and patients in G1 received full fat milk at 20 minutes post tracer injection, whereas patients in G2 received no intervention. Generally, routine imaging for stress is carried out 30 – 45 minutes post tracer injection, however in this study some patients were imaged later due to the longer acquisition times with the addition of prone imaging, which is a routine protocol in our department. After the rest injection of 740MBq of Tc-99m Sestamibi, patients in G0 received diluted lemon juice and patients in G1 received full fat milk, immediately post tracer injection, whereas patients in G2 had no intervention. The routine rest images were acquired 45 - 60 minutes post injection. All patients were imaged supine with their arms raised. Gated prone images were acquired thereafter.

2.4 Imaging protocol and Data Processing

SPECT imaging was performed using a double-head rotating large field of view Gamma camera (GE medical systems Infinia hybrid system), equipped with a low energy high resolution collimator. SPECT images were acquired on a 64 x 64 matrix. Sixty images (25 seconds for rest, 20 seconds for stress) were obtained over a semi-circular 180° arc. Filtered backprojection was performed with a low resolution Butterworth filter and no attenuation or scatter correction was applied. Transaxial tomograms were reconstructed and the images were re-orientated into three sets of orthogonal slices, including short – axis, horizontal long axis and vertical long axis for each study.

3.0 DATA ANALYSIS

Two experienced Nuclear Medicine physicians evaluated the raw data of the anterior (ANT) and left lateral (LLAT) views of both the stress and rest studies of all study participants for the presence or absence of interfering infra-cardiac activity. Slice numbers 15 and 45 of the planar display from the SPECT acquisition were used in all patients to increase reproducibility. Observers evaluated the images simultaneously and were blinded to the clinical information as well as the protocol details. If there was a disagreement with the values obtained a consensus was reached.

Visual and semi-quantitative assessment of the raw data of both stress and rest images as previously used by Hofman *et al* was used. [6] Visually, any presence of infra-cardiac activity was graded as 'yes' and the absence of infra-cardiac activity was graded as 'no'. If the infra-cardiac activity was equal to lung background it was described as absent. If infra-cardiac activity was present, it was graded as follows:

0: absence of infra-cardiac activity

1: infra-cardiac activity less than myocardial activity

2: infra-cardiac activity equal to myocardial activity

3: infra-cardiac activity greater than myocardial activity

For the semi-quantitative assessment, the total counts for the region of interest (ROI), which was manually drawn (six pixels wide), was obtained on an anterior static image (slice 15). The same ROI was copied to the infra-cardiac area below the inferior wall of the left ventricle. On the same raw data, the images were rotated to a lateral view (slice 45) and the ROI was copied to the inferior wall and the corresponding infra-cardiac area (Figures 1 & 2). Regions of interest (ROIs) were copied between stress and rest studies of individual patients to increase reproducibility.

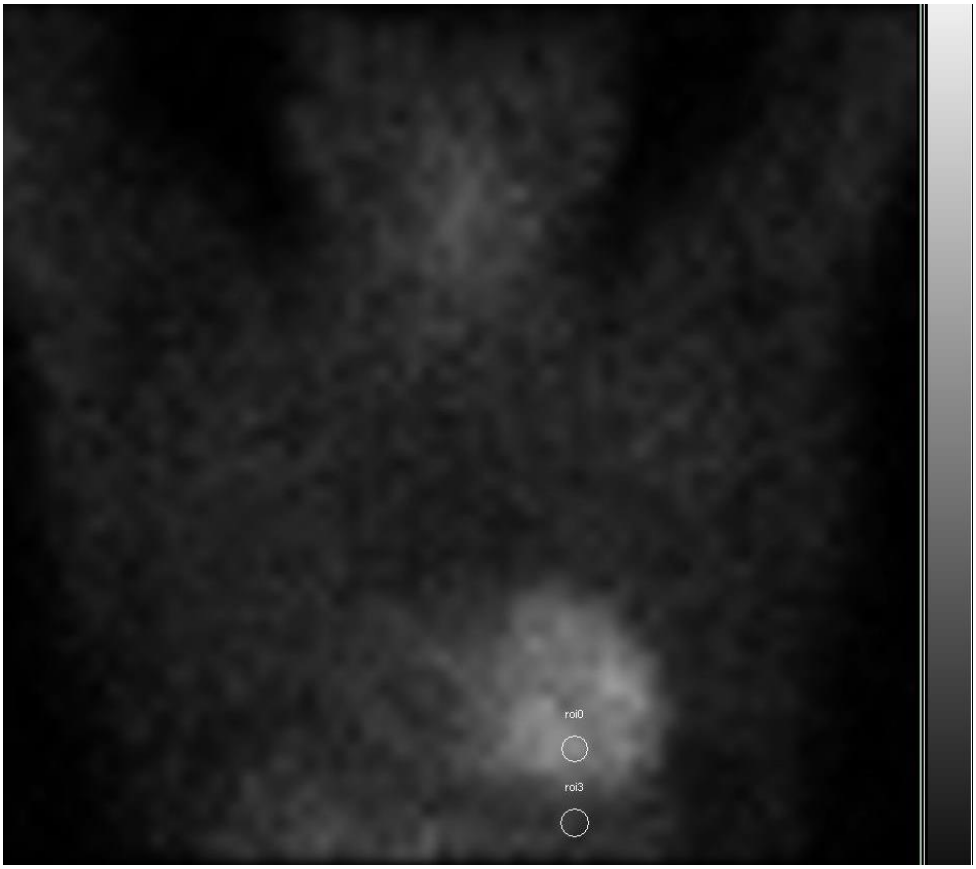

Figure 1: Anterior image. ROI in the inferior wall of the left ventricle copied to the infra-cardiac ROI

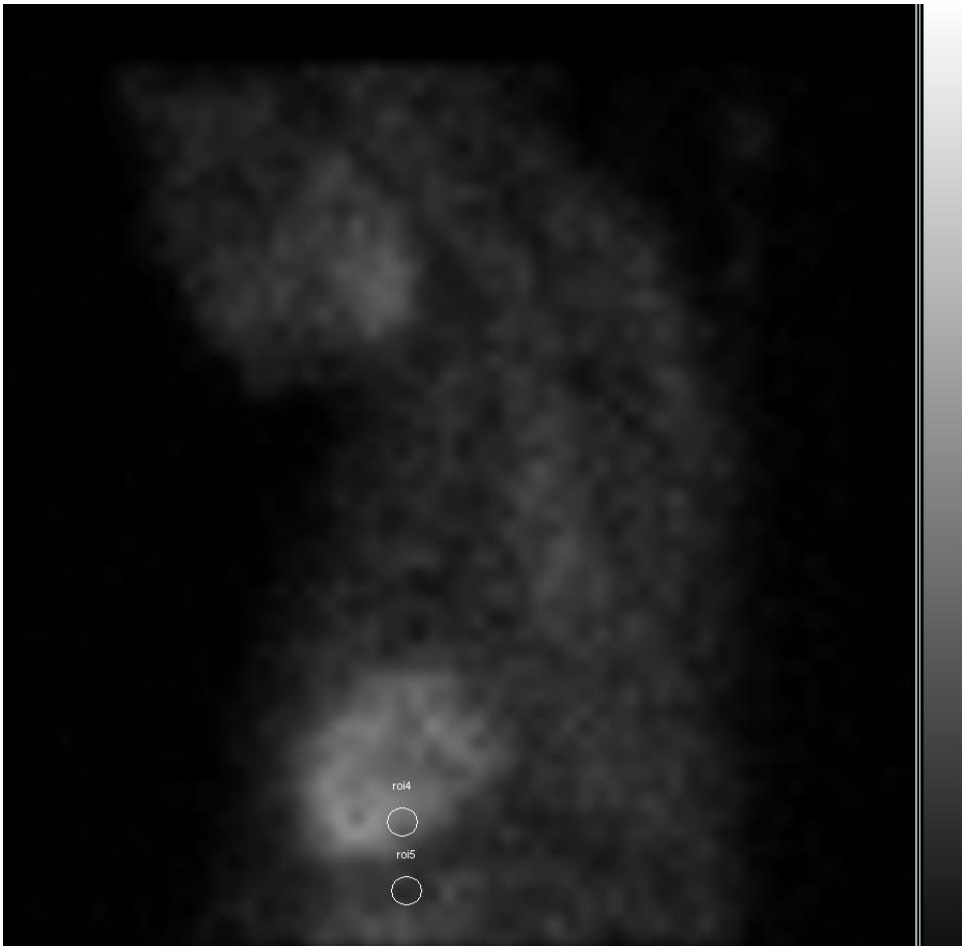

Figure 2: Lateral image. ROI in the inferior wall of the left ventricle copied to the infra-cardiac ROI

4.0 STATISTICAL ANALYSIS

Data were analyzed using a Statistica 10 package (statsoft Inc, Tilsa, Oklahoma, USA). [16]

Descriptive results were presented as medians and range (normal or not normally distributed) for continuous variables. Categorical variables were summarized as frequencies and percentages. To assess the differences between continuous variables (age, counts in the left ventricle and counts in the infra-cardiac region at rest and stress)[not normally distributed] a Kruskal – Wallis test was used between the three groups, (G0 ,G1 and G2), followed by the Bonferroni correction for two by two comparisons. Post hoc comparison of the mean ranks of all groups was performed. To compare frequencies of different categorical variables among the three groups, (G0, G1 and G2) the Chi – squared test or Fisher exact test was used when appropriate. Statistical significance was set at $p< 0.05$ and after Bonferroni correction at $p< 0.016$ for two by two comparisons.

5.0 RESULTS

Six hundred and thirty patients were randomized to receive either milk, lemon juice or no intervention and their characteristics are shown in table 2. Three hundred and thirteen patients received milk, two hundred and forty six patients received lemon juice and there was no intervention in seventy one patients. There were 304 females (48%) and 326 males (52%). The method of stress was exercise in 51% and pharmacological in 49% of patients. There was a statistically significant difference in the gender ($P=0.003$) and ethnicity ($P= 0.0002$) indexes among the different study groups.

Table 2: Patient characteristics

Characteristic	Total	Lemon juice (G0)	Milk (G1)	Control (G2)	P-value
N	630	246	313	71	
Age (year)[a]		58.21± 11.42	62.03± 11.43	61.37± 9.02	
Gender[b]					
Male	326(52)	109(44)	171(55)	46(65)	0.003
Female	304(48)	137(56)	142(45)	25(35)	
Stress[b]					
Exercise	319 (51)	127(52)	144(46)	48(68)	0.83
pharmacological	311(49)	119(48)	169(54)	23(32)	
Ethnicity[b]					
Black	193(30)	71(29)	81(26)	41(58)	
Caucasian	238(37)	95(39)	131(42)	12(17)	0.0002
Indian	140(22)	59(24)	68(22)	13(18)	
Coloured	59(11)	21(8)	33(10)	5(7)	

[a]: mean age and standard deviation
[b]: Values are represented as frequencies, and as percentages in parenthesis

In all three groups, the presence of infra-cardiac activity was present in the majority of patients both on the stress and rest studies (Table 3), however the grading (Table 4) made a significant difference in the groups with intervention, especially at rest and was more overt in G0, as compared to G2 (p= 0.013). At stress, majority of the patients in G0 and G1 had infra-cardiac activity less than or equal to myocardial activity. For the rest group, majority of patients in G0 and G1 had less or equal interfering infra-cardiac activity. It was interesting to note that just over one quarter of patients in G2 (27%) had infra-cardiac activity greater than myocardial activity as compared to G0 (19%) and G1 (18%).

The visual assessment for the presence or absence of infra-cardiac activity showed a statistically significant difference among the three groups, in post stress (p= 0.005) and at rest (P= 0.0063){Table 3}. The visual grading was statistically highly significant for the three groups, in post stress (p = 0.0002) and a similar difference was noted at rest (p = 0.004) {Table 4}

Table 3: Evaluation of infra-cardiac activity by visual assessment

	Total	Lemon juice group (G0)	Milk group (G1)	Control group (G2)	P - value
Stress[a]- presence of Infra-cardiac activity					0.005
Yes	528	201 (84.1)	257 (84.5)	70 (97)	
No	86	38 (15.9)	47 (15.5)	1 (3)	
Rest[a] – presence of Infra-cardiac activity					0.0063
Yes	564	219 (91.7)	274 (90.1)	71 (100)	
No	50	20 (8.3)	30 (9.9)	0	

Presence of infra-cardiac activity was graded as 'yes' and absence as 'no'

[a]: Values are represented as frequencies, and as percentages in parenthesis

Table 4: Visual grading of the intensity of infra-cardiac activity versus myocardial activity

Grading	0	1	2	3	P-value
Stress[a]					0.0002
Lemon juice (G0)	38(16)	138(58)	46(19)	17(7)	
Milk (G1)	47(16)	166(55)	49(16)	42(14)	
Control (G2)	1(1)	47(66)	19(28)	4(6)	
Rest[a]					0.004
Lemon juice (G0)	20(8)	100(42)	73(31)	46(19)	
Milk (G1)	29(10)	137(45)	84(28)	54(18)	
Control (G2)	0	24(34)	28(39)	19(27)	

Grading: 0: absent infra-cardiac activity; 1: infra-cardiac activity < myocardial activity; 2: infra-cardiac activity = myocardial activity; 3: infra-cardiac activity > myocardial activity.

[a] : Values are represented as frequencies, and as percentages in parenthesis

The analysis of the quantitative assessment of the total counts for all subjects and their comparisons within and between groups are given in table 5. The median was obtained in each group for the variable and their minimum and maximum values are included.

Table 5: Total counts in the myocardium and infra-cardiac area in the anterior and lateral positions

Region	Lemon juice (G0)	Milk (G1)	Control (G2)	Overall P value
S-inferior myocardium anterior[a]	634.5 (185-2648)*	733 (160-3260)#	553 (144-1566)	<0.0001
S-infra-cardiac anterior[a]	391 (79-1728)	429 (101-2551)#	364 (73-1308)	0.0106
S- inferior myocardium lateral[a]	584 (103-2100)*	673 (175-3913)#	534 (172-1693)	0.019
S-infra-cardiac lateral[a]	419 (63-2119)	452 (90-2347)	393 (88-1805)	0.1129
R- inferior myocardium anterior[a]	633.5 (186-8181)*	694 (36-2308)	586 (159-7171)	0.0089
R-infra-cardiac anterior[a]	464 (83-2101)	512 (89-2329)	443 (145-1288)	0.088
R- inferior myocardium lateral[a]	617.5 (109-2986)*	691 (32-2628)	612 (212-1897)	0.007
R-infra-cardiac lateral[a]	488.5 978-2672)*	552 (15-3037)	547 (126-1646)	0.020

S denotes stress image and R denotes rest image.

[a]: Values represent median of the total counts of a ROI. Values in parenthesis represent range.

*P <0.05 between the lemon juice group (G0) and the milk group (G1).

#p <0.05 between the milk group (G1) and the control group (G2).

Our findings are somewhat similar to the findings showing a decrease in infra-cardiac activity in accordance with earlier studies.[5,6,7,8,9,10,11,12,13] We have implemented the results of this study in our daily clinical practice with regard to milk administration. It is known that infra-cardiac activity is more common in rest myocardial perfusion images[2] as was shown in our study, therefore our current protocol for the rest MPI includes administration of 250mL full fat milk immediately after injection of the radiotracer.

6.0 DISCUSSION

In most African countries cardiovascular disease (CVD) is now the second commonest cause of death after infectious disease, accounting for 10% of total deaths and it is estimated that this burden will double from 1990 to 2020.[17,18] It also presents an enormous burden due to morbidity and health care expenses. MPI is a valuable tool in the management of patients with CVD and is currently used in Africa[19]. It has the ability to evaluate perfusion at a cellular level and at peak exercise stress. MPI plays an important role in diagnosing CVD, establishing prognosis, assessing the effectiveness of therapy and evaluating viability.

As mentioned earlier the use of Tc-99m Sestamibi for MPI often results in the increased splanchnic activity that may create a major problem in the visual and quantitative interpretation of inferior and infero-septal walls of the left ventricle. In fact, infra-cardiac activity arises predominantly from the liver, hepatobiliary system, bowel and/or gastro-duodenal reflux and can result in either an apparent increase or decrease in radiotracer uptake in the myocardium, especially in the inferior and infero-septal walls. [4]

Numerous studies have been carried out, using both Sestamibi and Tetrofosmin MPI, with various agents used to reduce infra-cardiac activity including, the oral administration of various fluids or solid meals and the use of pharmacologic agents. [5,6,7,8,9,10,11,12,13] The proposed mechanism of action is to fill the stomach, increasing the distance between the left ventricle and interfering infra-cardiac activity or to increase liver clearance of radiotracer via gallbladder contraction.

Among the studies performed using Tc-99m Sestamibi, *Vorster et al* [10] randomized 96 patients into two groups. Patients in group A received 500mg of non-enterically coated oral erythromycin 1 hour prior to image acquisition (45 patients) and patients in group B received 250mL diluted lemon juice (150mL lemon juice + 100mL water) ten minutes after tracer injection (51 patients). Differences between the

two groups varied between 6 and 10%. Interference was consistently higher on the stress studies when compared with the rest studies (p= 0.057 at stress and 0.0002 at rest). The presence of interfering infra-cardiac activity was consistently higher in the erythromycin group (55.56%) when compared with the lemon juice group (46.15%). However, the difference between the erythromycin and lemon juice group on interfering infra-cardiac activity was statistically insignificant, p= 0.36.

Hofman et al [6] compared milk versus water in reducing infra-cardiac activity in Tc-99m Sestamibi MPI. He randomized 198 patients into two groups. One group had 150mL chilled water and the other group had 150mL milk, 5 minutes after completion of the stress and again at 5 minutes before image acquisition. Patients also received 150mL chilled water or milk at 5 minutes after the rest injection and again at 5 minutes prior to the image acquisition (total 600mL of fluids for stress and rest images). There was a significant decrease in the intensity of infra-cardiac activity with milk compared to water. However the reduction in the intensity of infra-cardiac activity in the milk or water group did not translate into a statistically significant benefit in the image (p= 0.563 at stress and p= 0.502 at rest).

Malhotra et al [11] looked at the effect of carbonated lime drink prior to MPI with Tc-99m MIBI. There were a total of 33 patients that were injected with MIBI after the stress/rest. At 5 – 10 minutes post injection an anterior and left anterior oblique (LAO) was acquired. Thereafter the patients were given 250mL of a carbonated lime drink (40mL freshly squeezed limes and 210mL carbonated water) and repeat views were acquired within 5 minutes with the same parameters. It was found that the inferior wall was better visualized in the post-intervention planar and SPECT reconstructed views as compared with the pre-intervention images in all patients.

In a study of 260 patients, *Peace et al* [8] imaged patients at 0.5h, 1h or 2h post-injection and found that infra-cardiac activity significantly decreased with time ($p < 0.05$). The study also examined the effect on a subgroup of patients given 150mL full fat milk immediately after injection with a group given 150mL of

milk and 450mL of water between injection and imaging. There was no statistically significant difference in either group ($p > 0.05$).

Iqbal et al[12] proposed that iodinated oral contrast absorbs gamma rays and would therefore absorb scatter. Thirty patients were randomized undergoing adenosine Tc-99m Sestamibi to receive either one liter of iodinated oral contrast, one liter of water or no intervention. The improvement in image contrast was significant in the iodinated oral contrast and water group, ($p < 0.001$), but no statistically significant difference between the two interventions were seen.

In a study using Tc-99m Tetrofosmin, *Boz et al* [13] investigated the effect of a standardized meal of solid food and liquid (sandwich + 200mL water), in 60 patients undergoing exercise stress and found a reduction in the frequency of intestinal activity by visual assessment from 63% to 10%, ($p < 0.0001$) in the meal group and an increase in the inferior wall to abdomen count ratio of 1.48 to 2.49 ($p < 0.0001$) compared with no change in the control group.

All these studies have provided strong evidence that the administration of food or fluid results in a reduction of infra-cardiac activity. This occurs by pushing the sub-diaphragmatic activity caudally by increasing the distance between the inferior myocardial wall and the liver or bowel.

Similar results were noted in our study for patients referred for MPI in our local environment. However, our study had the largest number of patients (630) as compared to published studies in the literature. We had more than double the number of patients when compared to a study with the largest number of patients by *Peace et al.*[8]

6.1 Study Limitations

By excluding almost a third of the recruited patients (274) from the original number (904), the powers in each group were somewhat not the same.

7.0 CONCLUSION

Infra-cardiac activity causes significant artifacts after reconstruction and can lead to errors in visual and quantitative assessment of myocardial perfusion, especially in the inferior and infero-septal walls. Our study demonstrated that the administration of milk or lemon juice results in a significant decrease in the intensity of infra-cardiac activity as compared to the group with no intervention. At stress, both the milk group and the lemon juice group were comparable. At rest, the milk group showed a trend toward greater improvement than the group that received lemon juice. However, we are not sure on the impact of the amount of either milk or lemon juice as well as their timing of administration. Therefore, further studies are needed to determine the ideal amount and timing for administration of these interventions.

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9.0 APPENDIX

9.1 Tc-99m Sestamibi protocol for stress and rest imaging

Date: _____

Study protocol: 1 day ☐ 2 day ☐

Patient number: _____

Age: _____ Gender: _____ Weight:_____ Race: _____

Time of tracer injection: _____

Time of imaging: _____

Was prone imaging done? _____

Stress study ☐ (Physical or Pharmacological) rest study ☐

250ml milk ☐ lemon juice 50mL + 200mL water ☐ control ☐

(20 min post stress injection / immediately post rest injection)

Time of milk or juice administration: _____

Exclusion criteria:

- **previous cholecystectomy**
- **Liver/biliary system disease**
- **Peptic ulcer disease < 6 months**
- **Diabetes mellitus**
- **Myocardial infarct < 2 months**
- **Unstable angina**
- **Severe primary valvular disease**
- **Left ventricular hypertrophy**
- **LV aneurysm**
- **Primary cardiomegaly**
- **Severe conduction disturbances**
- **Lactose intolerance**

9.2 TITLE OF STUDY: EVALUATION OF THE EFFICACY OF FULL FAT MILK AND DILUTED LEMON JUICE VERSUS NO INTERVENTION TO REDUCE INTERFERING INFRA-CARDIAC ACTIVITY OF TC-99M SESTAMIBI DURING MYOCARDIAL PERFUSION IMAGING

Participant information sheet

Dear patient,

Good Morning,
Dr K Purbhoo of the Nuclear Medicine department at the CM Johannesburg and CH Baragwanath hospitals would like to invite you to join this study. Before you agree to take part in this study you should fully understand what is involved. If you have any questions, which are not fully explained in this information sheet, do not hesitate to ask the investigator. You should not agree to take part unless you are completely happy about the procedure involved.

The purpose of this study is to assess if giving you lemon juice or milk would result in better quality images/pictures when we report your scan. The research involves giving you a cup of milk or lemon juice to drink 20 minutes after we have given you the radiotracer injection. You should not experience any side-effects as a result of drinking the milk or lemon juice. You will later be put on a machine that takes images of your heart. This research will not affect your management in any way. Data obtained from this study may help in improving the results given to your cardiologist and therefore result in better management decisions when treating you.

No risk whatsoever is attached with this routine procedure. In our institution, we perform this procedure 40 to 60 times a week, for the detection and risk assessment of patients with suspected and known to have coronary artery disease.

There will be no cost to you by accepting to participate in this study. The direct benefit to you from this research would be to improve the quality of our reporting so that the cardiologists in our hospital can decide on the best way to treat you. Should you decline your consent, your treatment will not be affected and your decision will not influence your future care and continued treatment within this hospital.

A minimum of 500 patients will be enrolled in the study. All information obtained during the course of this research is strictly confidential. Data that may be reported in scientific journals will not include any information, which identifies you as a patient in this research. The results of this study may be submitted to the (HREC), Human Research Ethics Committee at their request.
Their contact details are as follows: telephone 011 717 1234, fax 011 717 1265 and e-mail Ms Anisa Keshav who is the secretary, anisa.keshav@wits.ac.za

Any information uncovered regarding your imaging results or state of health as a result of your participation in this trial will be held in strict confidence.
This study has been approved by the University of the Witwatersrand's human research ethics committee.
Your participation to this study is voluntary and should you have any questions regarding this study, please do not hesitate to contact Dr K Purbhoo, the telephone number is 011 488 3500 or 488 3590. My after hours contact number is 072 231 2548

Thank you for your time.

Written Consent Form

TITLE OF STUDY: EVALUATION OF THE EFFICACY OF FULL FAT MILK AND DILUTED LEMON JUICE VERSUS NO INTERVENTION TO REDUCE INTERFERING INFRA-CARDIAC ACTIVITY OF TC-99M SESTAMIBI DURING MYOCARDIAL PERFUSION IMAGING

Name of Patient:……

Patient Number:……

The aims and procedures of this study have been explained to me by the doctor. I have read and understood the subject information sheet provided.

I have had the opportunity to ask questions and to consider the answers given to me.

I understand that participation in this study is voluntary, that I may decline my consent and if I choose not to participate my decision will not affect my care and future visits at the hospital.

I hereby freely give my informed consent to taking part in this study.

NAME: ……..

DATE: ……

SIGNATURE: ……..

I confirm that I have fully explained the nature of the study and the procedure to be performed to the above named patient.

NAME: …….

DATE: ……..

SIGNATURE: …….

9.3 Evaluation of the efficacy of full fat milk, diluted lemon juice and no intervention to reduce interfering infra-cardiac activity of Tc-99m Sestamibi during Myocardial Perfusion Imaging

KHUSHICA PURBHOO, MDTHW VANGU

Summary

Objectives: When using Tc-99m Sestamibi for myocardial perfusion imaging, the increased infra-cardiac activity may create a major problem in the visual and quantitative interpretation of the inferior and infero-septal walls of the left ventricle. The aim of this study was to determine which protocol would be more effective to reduce interfering infra-cardiac activity and therefore result in an improvement in image quality. We compared the use of full fat milk and of diluted lemon juice to no intervention.

Methods: All patients referred to our institution for myocardial perfusion imaging from November 2009 to May 2012 were enrolled in the study. A total of 630 patients were randomized without stratification into three groups. Group 0 (G0) were given diluted lemon juice, 246 patients, full fat milk to group 1 (G1), 313 patients and group 2 (G2), 71 patients, had no intervention. The latter was the control group. A routine two day protocol was used and the patients were given the same intervention on both days. Raw data of both the stress and rest images were visually and quantitatively assessed by two Nuclear Medicine physicians for the presence of infra-cardiac activity. The physicians were blinded to the intervention received and the data were reviewed simultaneously.

Results: The overall incidence of interfering infra-cardiac activity at stress was 84.1%, 84.5% and 96.6% in G0, G1 and G2, respectively (P= 0.005) and at rest was 91.7%, 90.1% and 100% in G0, G1 and G2, respectively (P= 0.0063). The visual and quantitative results favoured both milk and lemon juice in reducing the amount of interfering infra-cardiac activity.

Conclusion: The administration of milk or lemon juice resulted in a significant decrease in the intensity of infra-cardiac activity compared to the control group. This reduction in intensity was even more significant in the milk group for patients done during rest myocardial perfusion imaging.

Keywords: myocardial perfusion imaging, full fat milk, lemon juice, infra-cardiac activity, Sestamibi, Tc-99m

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Introduction

Coronary artery disease is one of the major causes of death throughout the world.

The use of Single Photon Emission Computed Tomography (SPECT) myocardial perfusion imaging (MPI), with technetium 99m labeled radiopharmaceuticals [Tc-99m Sestamibi (Methoxy isobutyl isonitrile) and Tc-99m Tetrofosmin] in conjunction with either exercise or pharmacologic stress is an established tool for both the diagnosis and prognostication of patients with ischemic heart disease.[1]

The basis for the non-invasive approach is that physiologic alterations of regional myocardial blood flow or systolic contraction of the myocardium caused by stress may be more predictive of outcome than the knowledge of coronary anatomy alone. Patients with normal perfusion on Tc-99m SPECT MPI have an excellent prognosis, whereas patients with abnormal scans have an increased rate of cardiac death and non-fatal infarction during follow-up.[2]

For perfusion imaging with SPECT, Thallium 201 (Tl-201) and Tc-99m labeled radiopharmaceuticals are commonly used. The major metabolic pathway for clearance of Sestamibi is the hepatobiliary system, thereby infra-cardiac activity from the liver and bowel may impact on the interpretation of the inferior wall during reconstruction. Activity can also be present in the stomach due to reflux of tracer into the gastric lumen from the duodenum or because of uptake of free pertechnetate by the gastric mucosa. The presence of infra-cardiac activity leads to artifacts, reducing the desired target to background ratio creating difficulty in both visual and quantitative interpretation of myocardial perfusion, particularly in the inferior and infero-septal walls.[2]

Infra-cardiac activity is less common with exercise and is more common with pharmacologic stress and/or in rest studies.[3]

Several different protocols including, a fatty meal, milk, milk and water, lemon juice, milkshake, carbonated drinks, iodinated oral contrast, intravenous injection of cholecystokinin and the

administration of metoclopramide or erythromycin have been described as means to reduce the artifacts arising from abdominal activity.[4-13] Depending on the protocol used the mechanisms for these interventions will include one or more of the following:

- Expansion of the stomach by the volume effect, displacing it caudally thereby increasing the distance between the heart and the intestine
- Increased gastric emptying
- Stimulation of liver clearance and peristaltic movement
- Acceleration of bile secretion and gallbladder emptying

Patients and methods

Study population

All patients 18 years and older who were referred for MPI were enrolled in the study. Ethics approval was obtained from the University of the Witwatersrand's Human Research Ethics Committee and written consent was obtained from all study participants. The study commenced in November 2009 until May 2012. We recruited 904 patients in this study but data from 274 patients were excluded due to various reasons [non-return for second day study, milk or lemon juice not followed in a patient for both the stress and rest study and patients who fitted the exclusion criteria (Table 1)]. A total of 630 patients (304 female [48%] and 326 male [52%]), aged 19 – 84 years were eventually enrolled for data analysis.

Randomization

Patients were randomized into three groups. Group 0 (G0) had diluted lemon juice, group 1 (G1) had full fat milk and group 2 (G2) had no intervention (control group). Full fat milk consisted of 250mL milk. Diluted lemon juice consisted of 50mL lemon juice and 200mL water, total volume of 250mL.

Patient preparation and Stress protocol

A routine two day protocol was used. Patients were stressed on day one and a rest study was done on day two. Patients were fasted at least four hours prior to stress testing (usually overnight) and were required to abstain from caffeine- containing beverages and methylxanthine containing medications for at least 24 hours. The reason is that caffeine and methylxanthines block the adenosine receptors on arterial smooth muscle cells, thus limiting the effectiveness of vasodilator agents. Our department protocol is that we withhold caffeine in all patients even if exercise stress is planned in case there is a necessity to switch to pharmacological stress. Beta-blockers and calcium channel antagonists were also withheld. The patients were hemodynamically and clinically stable for 48 hours prior to the test.

The stress modality (Treadmill, Dipyridamole or Dobutamine) was chosen and implemented in accordance with the recent EANM guideline.[14] Following the injection of 740MBq Tc-99m Sestamibi during stress, patients in G0 received diluted lemon juice and patients in G1 received full fat milk at 20 minutes post tracer injection, whereas patients in G2 received no intervention. Generally routine imaging for stress is carried out 30 – 45 minutes post tracer injection, however in this study some patients were imaged later due to the longer acquisition times with the addition of prone imaging, which is also a routine protocol in our department. After the rest injection of 740MBq of Tc-99m

Sestamibi, patients in G0 received diluted lemon juice and patients in G1 received full fat milk, immediately post tracer injection, whereas patients in G2 received no intervention. The routine rest images were acquired 45 - 60 minutes post injection. All patients were imaged supine with their arms raised. Gated prone images were acquired thereafter for the stress images.

Imaging protocol and Data Processing

SPECT imaging was performed using a double-head rotating large field of view Gamma camera (GE medical systems Infinia hybrid system), equipped with a low energy high resolution collimator. SPECT images were acquired on a 64 x 64 matrix. Sixty images (25 seconds for rest, 20 seconds for stress) were obtained over a semi-circular 180° arc. Filtered backprojection was performed with a low resolution Butterworth filter and no attenuation or scatter correction was applied. Transaxial tomograms were reconstructed and the images were re-orientated into three sets of orthogonal slices, including short – axis, horizontal long axis and vertical long axis for each study.

Data analysis

Two experienced Nuclear medicine physicians evaluated the raw data of the anterior (ANT) and left lateral (LLAT) views of both the stress and rest studies of all study participants for the presence or absence of interfering infra-cardiac activity. Slice numbers 15 and 45 of the planar display from the SPECT acquisition were used in all patients to increase reproducibility. Observers evaluated the images simultaneously and were blinded to the clinical information as well as the protocol details. If there was a disagreement with the values obtained a consensus was reached.

The observers used visual and semi-quantitative assessment of the raw data of both stress and rest images as previously used by Hofman *et al.*[5] Visually, any presence of infra-cardiac activity was graded as

'yes' and the absence of infra-cardiac activity was graded as 'no'. If the infra-cardiac activity was equal to lung background it was described as absent. If infra-cardiac activity was present, it was graded as follows:

0: absence of infra-cardiac activity

1: infra-cardiac activity less than myocardial activity

2: infra-cardiac activity equal to myocardial activity

3: infra-cardiac activity greater than myocardial activity

For the semi-quantitative assessment, the total counts for the region of interest (ROI), which was manually drawn (six pixels wide), was obtained on an anterior static image (slice 15). The same ROI was copied to the infra-cardiac area below the inferior wall of the left ventricle. On the same raw data, the images were rotated to a lateral view (slice 45) and the ROI was copied to the inferior wall and the corresponding infra-cardiac area (Figures 1 & 2). Regions of interest (ROIs) were copied between stress and rest studies of individual patients to increase reproducibility.

Statistical analysis

Data were analyzed using a Statistica 10 package (statsoft Inc, Tilsa, Oklahoma, USA). [16]
Descriptive results were presented as medians and range (normal or not normally distributed) for continuous variables. Categorical variables were summarized as frequencies and percentages. To assess the differences between continuous variables (age, counts in the left ventricle and infra-cardiac region at rest and stress)[not normally distributed] a Kruskal – Wallis test was used between the three groups, (G0 ,G1 and G2), followed by the Bonferroni correction for two by two comparisons. Post hoc comparison of the mean ranks of all groups was performed. To compare frequencies of different

categorical varia bles among the three groups, (G0, G1 and G2) the Chi – squared test or Fisher exact test was used when appropriate. Statistical significance was set at $p< 0.05$ and after Bonferroni correction at $p< 0.016$ for two by two comparisons.

Results

Six hundred and thirty patients were randomized to receive either milk, lemon juice or no intervention and their characteristics are shown in table 2. Three hundred and thirteen patients received milk, two hundred and forty six patients received lemon juice and there was no intervention in seventy one patients. There were 304 females (48%) and 326 males (52%). The method of stress was exercise in 51% and pharmacological in 49% of patients. There was a statistically significant difference in the gender (P=0.003) and ethnicity (P=0.0002) indexes among the different study groups.

In all three groups, the presence of infra-cardiac activity was present in the majority of patients both on the stress and rest studies (Table 3), however the grading (Table 4) made a significant difference in the groups with intervention, especially at rest and was more overt in G0, as compared to G2 (p=0.013). At stress, the majority of the patients in G0 and G1 had infra-cardiac activity less than or equal to myocardial activity. For the rest group, the majority of patients in G0 and G1 had less or equal interfering infra-cardiac activity. It was interesting to note that just over one quarter of patients in G2 (27%) had infra-cardiac activity greater than myocardial activity as compared to G0 (19%) and G1 (18%).

The visual assessment for the presence or absence of infra-cardiac activity showed a statistically significant difference among the three groups, in post stress (p=0.005) and at rest (p=0.0063)[Table 3].

The difference in visual grading was also statistically highly significant for the three groups, in post stress ($p = 0.0002$) and a similar difference was noted at rest ($p = 0.004$) {Table 4}.

The analysis of the quantitative assessment of the total counts for all subjects and their comparisons within and between groups are given in table 5. The median was obtained in each group for the variable and their minimum and maximum values are included.

Our findings are overall in accordance with earlier studies in showing a decrease in infra-cardiac activity.[4,5,8,9,10,12,13,14] We have implemented the results of this study in our daily clinical practice with regard to milk administration. It is known that infra-cardiac activity is more common in rest myocardial perfusion images[2] as was shown in our study, therefore our current protocol for the rest MPI includes administration of 250mL full fat milk immediately after injection of the radiotracer.

Discussion

In most African countries cardiovascular disease (CVD) is now the second commonest cause of death after infectious disease, accounting for 10% of total deaths and it is estimated that this burden will double from 1990 to 2020.[16,17] It also presents an enormous burden due to morbidity and health care expenses. MPI is a valuable tool in the management of patients with CVD and is currently used in Africa[18]. It has the ability to evaluate perfusion at a cellular level and at peak exercise stress. MPI plays an important role in diagnosing CVD, establishing prognosis, assessing the effectiveness of therapy and evaluating viability.

As mentioned earlier the use of Tc-99m Sestamibi for MPI often results in increased splanchnic activity that may create a major problem in the visual and quantitative interpretation of inferior and infero-septal walls of the left ventricle. In fact, infra-cardiac activity arises predominantly from the liver,

hepatobiliary system, bowel and/or gastro-duodenal reflux and can result in either an apparent increase or decrease in radiotracer uptake in the myocardium, especially in the inferior and infero-septal walls. [4]

Numerous studies have been carried out, using both Sestamibi and Tetrofosmin MPI, with various agents used to reduce infra-cardiac activity, including the oral administration of various fluids or solid meals and the use of pharmacologic agents. [5,6,7,8,9,10,11,12,13] The proposed mechanism of action is to fill the stomach, increasing the distance between the left ventricle and interfering infra-cardiac activity or to increase liver clearance of radiotracer via gallbladder contraction.

Among the studies performed using Tc-99m Sestamibi, *Vorster et al* [10] randomized 96 patients into two groups. Patients in group A received 500mg of non-enterically coated oral erythromycin 1 hour prior to image acquisition (45 patients) and patients in group B received 250mL diluted lemon juice (150mL lemon juice + 100mL water) ten minutes after tracer injection (51 patients). Differences between the two groups varied between 6 and 10%. Interference was consistently higher on the stress studies when compared with the rest studies (p=0.057 at stress and 0.0002 at rest). The presence of interfering infra-cardiac activity was consistently higher in the erythromycin group (55.56%) when compared with the lemon juice group (46.15%). However, the difference between the erythromycin and lemon juice group on interfering infra-cardiac activity was statistically insignificant, p= 0.36.

Michael et al [5] compared milk versus water in reducing infra-cardiac activity in Tc-99m Sestamibi MPI. He randomized 198 patients into two groups. One group had 150mL chilled water and the other group had 150mL milk, 5 minutes after completion of the stress and again at 5 minutes before image acquisition. Patients also received 150mL chilled water or milk at 5 minutes after the rest injection and again at 5 minutes prior to the image acquisition (total 600mL of fluids for stress and rest images). There was a significant decrease in the intensity of infra-cardiac activity with milk compared to water.

However the reduction in the intensity of infra-cardiac activity in the milk or water group did not translate into a statistically significant benefit in the image (p=0.563 at stress and p=0.502 at rest).

Malhotra et al [11] looked at the effect of carbonated lime drink prior to MPI with Tc-99m MIBI. There were a total of 33 patients that were injected with MIBI after the stress/rest. At 5 – 10 minutes post injection an anterior and left anterior oblique (LAO) was acquired. Thereafter the patients were given 250mL of a carbonated lime drink (40mL freshly squeezed limes and 210mL carbonated water) and repeat views were acquired within 5 minutes with the same parameters. It was found that the inferior wall was better visualized in the post-intervention planar and SPECT reconstructed views as compared with the pre-intervention images in all patients.

In a study of 260 patients, *Peace et al* [8] imaged patients at 0.5h, 1h or 2h post-injection and found that infra-cardiac activity significantly decreased with time ($p<0.05$). The study also examined the effect on a subgroup of patients given 150mL full fat milk immediately after injection with a group given 150mL of milk and 450mL of water between injection and imaging. There was no statistically significant difference in either group ($p>0.05$).

Iqbal et al[12] proposed that iodinated oral contrast absorbs gamma rays and would therefore absorb scatter. Thirty patients were randomized undergoing adenosine Tc-99m Sestamibi stress MPI to receive either one liter of iodinated oral contrast, one liter of water or no intervention. The improvement in image contrast was significant in the iodinated oral contrast and water group, ($p< 0.001$), but no statistically significant difference between the two interventions were seen.

In a study using Tc-99m Tetrofosmin, *Boz et al* [13] investigated the effect of a standardized meal of solid food and liquid (sandwich + 200mL water), in 60 patients undergoing exercise stress and found a reduction in the frequency of intestinal activity by visual assessment from 63% to 10%, ($p <0.0001$) in

the meal group and an increase in the inferior wall to abdomen count ratio of 1.48 to 2.49 ($p < 0.0001$) compared with no change in the control group.

All these studies have provided strong evidence that the administration of food or fluid results in a reduction of infra-cardiac activity. As previously mentioned, this occurs by pushing the sub-diaphragmatic activity caudally by increasing the distance between the inferior myocardial wall and the liver or bowel.

Similar results were noted in our study for patients referred for MPI in our local environment. However, our study had the largest number of patients (630) as compared to published studies in the literature. We had more than double the number of patients when compared to a study with the largest number of patients by *Peace et al.*[8]

Study Limitations

By excluding almost a third of the recruited patients (274) from the original number (904), the powers in each group were not the same.

Conclusion

Infra-cardiac activity causes significant artifacts after reconstruction and can lead to errors in visual and quantitative assessment of myocardial perfusion, especially in the inferior and infero-septal walls. Our study demonstrated that the administration of milk or lemon juice results in a significant decrease in the intensity of infra-cardiac activity as compared to the group with no intervention. At stress, both the milk group and the lemon juice group were comparable. At rest, the milk group showed a trend toward greater improvement than the group that received lemon juice. However, we are not sure on the impact

of the amount of either milk or lemon juice as well as their timing of administration. Therefore, further studies are needed to determine the ideal amount and timing for administration of these interventions.

Acknowledgements

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Appendices

Table 1. Inclusion and exclusion criteria

Inclusion Criteria	Exclusion Criteria
• Patients older than 18 years of age • Patients referred for Tc-99m Sestamibi Myocardial Perfusion scans	• Lactose intolerance • Patients who failed exercise and had a contraindication to pharmacologic stress testing ie. Vasodilators and dobutamine • Inability to drink 250 ml of fluids secondary to medically essential fluid restriction • Pregnancy • Previous cholecystectomy, liver or biliary system disease • Peptic ulcer disease within the last 6 months • History of diabetes mellitus • Previous myocardial infarction within the last 2 months, unstable angina, severe primary valvular disease, left ventricular aneurysm, primary cardiomegaly, left ventricle hypertrophy or severe conduction disturbances

Characteristic	Total	Lemon juice (G0)	Milk (G1)	Control (G2)	P-value
N	630	246	313	71	
Age (year)[a]		58.21± 11.42	62.03± 11.43	61.37± 9.02	
Gender[b]					
Male	326(52)	109(44)	171(55)	46(65)	0.003
Female	304(48)	137(56)	142(45)	25(35)	
Stress[b]					
Exercise	319 (51)	127(52)	144(46)	48(68)	0.83
pharmacological	311(49)	119(48)	169(54)	23(32)	
Ethnicity[b]					
Black	193(30)	71(29)	81(26)	41(58)	
Caucasian	238(37)	95(39)	131(42)	12(17)	0.0002
Indian	140(22)	59(24)	68(22)	13(18)	
Coloured	59(11)	21(8)	33(10)	5(7)	

Table 2: Patient characteristics

[a]: mean age and standard deviation

[b]: Values are represented as frequencies, and as percentages in parenthesis

	Total	Lemon juice group (G0)	Milk group (G1)	Control group (G2)	P - value
Stress[a]- presence of Infra-cardiac activity					0.005
Yes	528	201 (84.1)	257 (84.5)	70 (97)	
No	86	38 (15.9)	47 (15.5)	1 (3)	
Rest[a] – presence of Infra-cardiac activity					0.0063
Yes	564	219 (91.7)	274 (90.1)	71 (100)	
No	50	20 (8.3)	30 (9.9)	0	

Table 3: Evaluation of infra-cardiac activity by visual assessment

Presence of infra-cardiac activity was graded as 'yes' and absence as 'no'

[a]: Values are represented as frequencies, and as percentages in parenthesis

Grading	0	1	2	3	P-value
Stress[a]					0.0002
Lemon juice (G0)	38(16)	138(58)	46(19)	17(7)	
Milk (G1)	47(16)	166(55)	49(16)	42(14)	
Control (G2)	1(1)	47(66)	19(28)	4(6)	
Rest[a]					0.004
Lemon juice (G0)	20(8)	100(42)	73(31)	46(19)	
Milk (G1)	29(10)	137(45)	84(28)	54(18)	
Control (G2)	0	24(34)	28(39)	19(27)	

Table 4: Visual grading of the intensity of infra-cardiac activity versus myocardial activity

Grading: 0: absent infra-cardiac activity; 1: Infra-cardiac activity < myocardial activity; 2: Infra-cardiac activity = myocardial activity; 3: Infra-cardiac activity > myocardial activity.

[a] : Values are represented as frequencies, and as percentages in parenthesis

Region	Lemon juice (G0)	Milk (G1)	Control (G2)	Overall P value
S-inferior myocardium anterior[a]	634.5 (185-2648)*	733 (160-3260)#	553 (144-1566)	<0.0001
S-infra-cardiac anterior[a]	391 (79-1728)	429 (101-2551)#	364 (73-1308)	0.0106
S- inferior myocardium lateral[a]	584 (103-2100)*	673 (175-3913)#	534 (172-1693)	0.019
S-infra-cardiac lateral[a]	419 (63-2119)	452 (90-2347)	393 (88-1805)	0.1129
R- inferior myocardium anterior[a]	633.5 (186-8181)*	694 (36-2308)	586 (159-7171)	0.0089
R-infra-cardiac anterior[a]	464 (83-2101)	512 (89-2329)	443 (145-1288)	0.088
R- inferior myocardium lateral[a]	617.5 (109-2986)*	691 (32-2628)	612 (212-1897)	0.007
R-infra-cardiac lateral[a]	488.5 978-2672)*	552 (15-3037)	547 (126-1646)	0.020

Table 5: Total counts in the myocardium and infra-cardiac area in the anterior and lateral positions
S denotes stress image and R denotes rest image.
[a]: Values represent median of the total counts of a ROI. Values in parenthesis represent range.
*P <0.05 between the lemon juice group (G0) and the milk group (G1).
#p <0.05 between the milk group (G1) and the control group (G2).

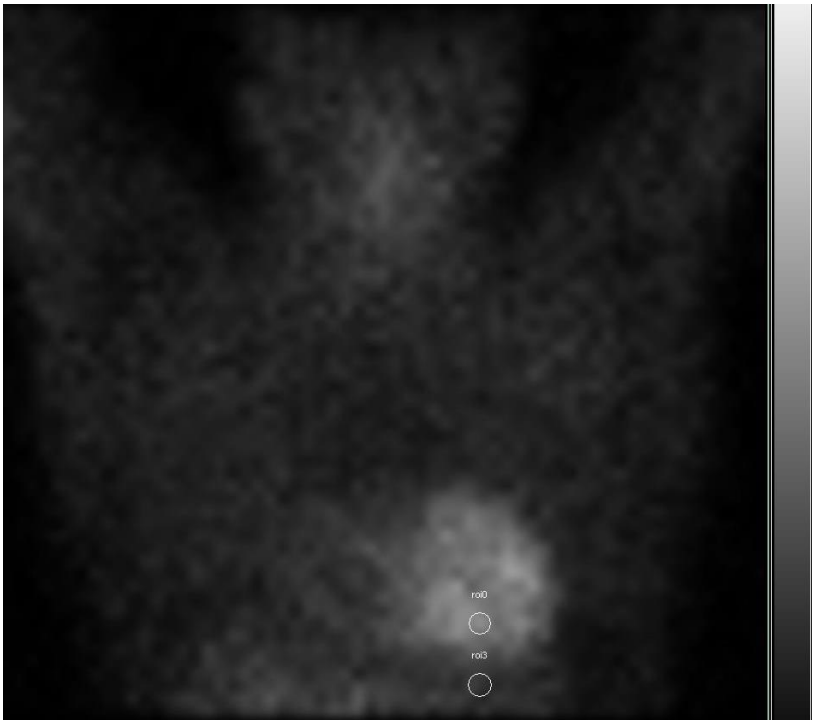

Figure 1: Anterior image. ROI in the inferior wall of the left ventricle copied to the infra-cardiac ROI

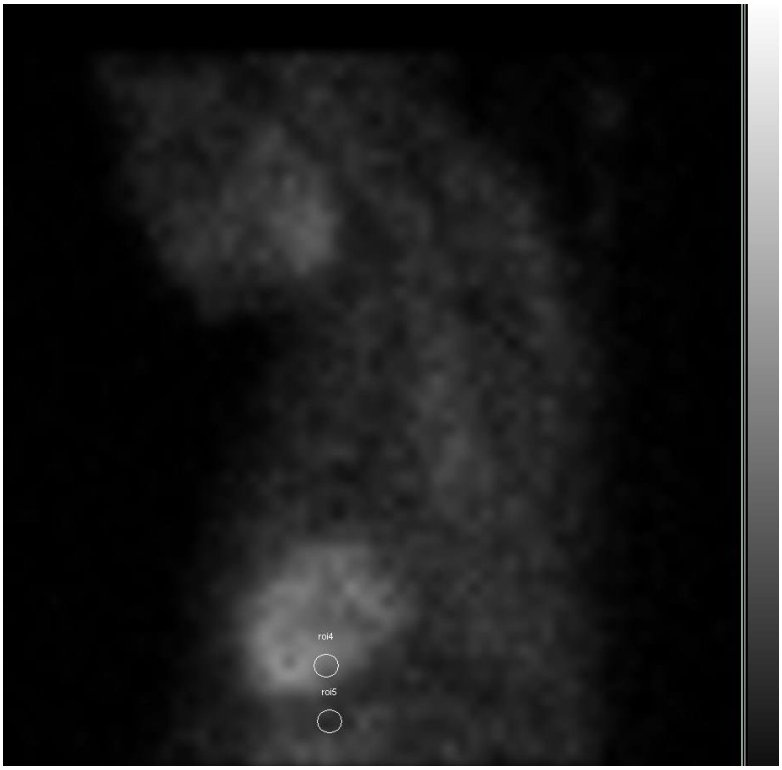

Figure 2: Lateral image. ROI in the inferior wall of the left ventricle copied to the infra-cardiac ROI

8.2 Ethics clearance

UNIVERSITY OF THE WITWATERSRAND, JOHANNESBURG

Division of the Deputy Registrar (Research)

HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)
R14/49 Dr Khushica Purbhoo

CLEARANCE CERTIFICATE	**M091012**
PROJECT	Efficacy of Full Fat verses Lemon Juice to Reduce Interfering Infra-cardiac Activity in TC-99M Sestamibi Myocardial Perfusion Imaging (MPI)
INVESTIGATORS	Dr Khushica Purbhoo.
DEPARTMENT	Department of Nuclear Medicine
DATE CONSIDERED	2009/10/30
DECISION OF THE COMMITTEE*	Approved unconditionally

Unless otherwise specified this ethical clearance is valid for 5 years and may be renewed upon application.

DATE 2009/11/02 **CHAIRPERSON** .....
(Professor PE Cleaton-Jones)

*Guidelines for written 'informed consent' attached where applicable

cc: Supervisor : Prof MDTHW Vangu

DECLARATION OF INVESTIGATOR(S)

To be completed in duplicate and **ONE COPY** returned to the Secretary at Room 10004, 10th Floor, Senate House, University.
I/We fully understand the conditions under which I am/we are authorized to carry out the abovementioned research and I/we guarantee to ensure compliance with these conditions. Should any departure to be contemplated from the research procedure as approved I/we undertake to resubmit the protocol to the Committee. **I agree to a completion of a yearly progress report.**

PLEASE QUOTE THE PROTOCOL NUMBER IN ALL ENQUIRIES...