An Evaluation of ^{99m}Tc-MIBI Imaging of Kaposi's Sarcoma in AIDS Patients

by

Fawzia Ismail Peer

Dissertation submitted in compliance with the requirement for the Degree of Doctorate of Technology: Radiography to the Department of Radiography, Durban Institute of Technology

December 2005

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I, Fawzia Ismail Peer, declare that this dissertation represents my own work, both in conception and execution and has not been submitted for any other qualification at any other institution

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^{99m}Tc-MIBI Imaging of AIDS-Associated Kaposi's Sarcoma in the Lungs.

by Fawzia I. Peer, Margaret H. Pui, William I. D. Rae, Anisa Mosam.

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LIST OF ABBREVIATIONS

^{99m}Tc	99m-Technetium
^{99m}Tc-MIBI	99m-Technetium-Methoxyisobutylisonitrile
^{99m}Tc-O_4^-	99m-Technetium- Pertecnetate
AIDS	Acquired Immunodeficiency Syndrome
ANOVA	Analysis of Variance
ART	Anti Retroviral Therapy
BAL	Broncheolar Lavage
FDG	Fluorodeoxyglucose
Ga	Gallium
GM	Geometric Mean
HHV	Human Herpes Virus
HIV	Human Immunodeficient Virus
IALCH	Inkosi Albert Luthuli Central Hospital
KS	Kaposi's Sarcoma
LLR	Local Linear Regression
LRR	Lung Retention Ratio
MDR	Multi Drug Resistant
Mo	Molybdenum
NPV	Negative Predictive Value
OIs	Opportunistic Infections
PCP	*Pneumocystis carinii* (*Pneumocystis jirovecii)* pneumonia
PET	Positron Emission Tomography

PPV	Positive Predictive Value
ROI	Region of Interest
SPECT	Single Photon Emission Computed Tomography
Tl	Thallium
TB	Tuberculosis
TBB	Transbronchial Biopsy

ABSTRACT

AIM

The purpose of this study was to evaluate ^{99m}Tc- methoxyisobutylisonitrile (MIBI) imaging, in terms of sensitivity and specificity, for non invasively detecting extracutaneous involvement of Kaposi's sarcoma (KS) and for differentiating pulmonary infection from malignancy in acquired immunodeficiency syndrome (AIDS) patients before and after treatment. Current investigations are invasive.

HYPOTHESIS

It was hypothesized that ^{99m}Tc-MIBI imaging would be both sensitive and specific for the detection of extracutaneous involvement of KS before and after treatment in AIDS patients and that ^{99m}Tc-MIBI imaging would be able to differentiate pulmonary infection from malignancy in AIDS patients.

METHOD

Using a non-randomized convenience sampling technique, sixty-six AIDS patients with biopsy-proven KS who met the inclusion criteria were studied in the sample cohort. The mean age of the patients in the sample was 33.6 years with the distribution of the males and females being 49% and 51% respectively. Every patient had a bronchoscopy and a series of ^{99m}Tc-MIBI scans prior to treatment. Follow-up ^{99m}Tc-MIBI scans were performed on the four patients that returned post-treatment.

Fifteen consenting non-KS AIDS patients with confirmed pulmonary pathology on bronchoscopy formed the control group of the study. Every patient had ^{99m}Tc-MIBI imaging using the same protocol as the sample group.

The investigator was blinded to the bronchoscopic findings. The scan results were quantified and various time delayed lung uptake ratios were obtained. The ^{99m}Tc-MIBI imaging results were compared to the bronchoscopy findings. The final patient diagnosis was made by histological, microbiological or bronchoscopic findings. Using the final diagnosis as the gold standard, sensitivity, specificity, and accuracy of the visual interpretation and quantitative analysis methods was determined. In order to meet the objectives of the study the results were divided into chest and cutaneous analysis.

Chest Analysis

The data obtained from the chest images were divided into four (4) groups,

i) KS patients with KS lung involvement

ii) KS patients with no KS lung involvement

iii) KS patients with other lung pathology

iv) Non-KS patients with other lung pathology

Lung uptake in KS and opportunistic infection were compared on planar and Single Photon Emission Computed Tomography (SPECT) scans.

Cutaneous Analysis

Assessment of cutaneous lesions on the whole body planar imaging was confined to the extremities because of normal radioactive uptake of ^{99m}Tc-MIBI in the salivary glands, pharynx, myocardium, hepatobiliary, gastrointestinal, and urinary tracts that can partly obscure abnormal uptake in the face and trunk.

RESULTS

The 60-minute lung/myocardium uptake ratios were significantly higher in KS and normal lungs than in opportunistic infection. Follow-up scans in patients post-treatment for KS showed a significant decrease in the 60-minute lung/myocardium uptake ratios.

Abnormal uptake of ^{99m}Tc-MIBI in the skin, subcutaneous tissue and lymph nodes was compared with the clinical assessment. Good correlation was noted between the clinical findings and the detection of cutaneous KS in the extremities using ^{99m}Tc-MIBI imaging. Abnormal lymph nodes and lymphoedema were detected in more patients on ^{99m}Tc-MIBI scans than on clinical assessment. SPECT was more effective than planar imaging for demonstrating abnormal lymph nodes, pericardial effusions and ascites. ^{99m}Tc-MIBI imaging provided additional information on the extent of lymph node involvement, which could be used for more precise staging and therapeutic planning of KS.

Cutaneous KS is often complicated by venous stasis of the extremities, face and genitals. Venous stasis was demonstrated proximal to the ^{99m}Tc-MIBI-injection site as dilated veins with persistent retention of radioactivity at 60 minutes post injection in more than 20% of the KS-patients. The reason for this observation is not certain. This is possibly the first report of an imaging finding of persistent venous activity in KS.

CONCLUSION

From the quantitative and qualitative assessments it is clear that ^{99m}Tc-MIBI imaging is not ideal for differentiating pulmonary KS from opportunistic infection in AIDS-KS patients, but could prove useful for differentiating pulmonary infection

from malignancy in non-KS-AIDS patients. ^{99m}Tc-MIBI imaging may be useful for follow-up of therapeutic response of KS.

Good correlation was seen between the clinical findings and detecting cutaneous KS in the extremities using ^{99m}Tc-MIBI imaging. ^{99m}Tc-MIBI imaging provides additional information on the extent of lymph node involvement and more precise staging and therapeutic planning. SPECT compared to planar imaging with ^{99m}Tc-MIBI was more effective for demonstrating abnormal lymph nodes, pericardial effusions and ascites. Clinical trials to test the efficacy of ^{99m}Tc-MIBI SPECT imaging of the abdomen and pelvis may be necessary.

The ^{99m}Tc-MIBI scans performed on patients post-treatment demonstrate the therapeutic effect in patients with no uptake of skin lesions and showed decreased lymph node uptake. The number of patients who could be followed up was small but these results indicate that ^{99m}Tc-MIBI imaging could prove useful as a predictive test or as a follow-up test to determine response of KS to treatment for extracutaneous involvement of KS.

RECOMMENDATION

Planar whole-body and SPECT ^{99m}Tc -MIBI imaging may be used as a non invasive diagnostic tool for the detection of extracutaneous involvement of KS as it provides additional information on the extent of lymph node involvement, and lymphoedema. ^{99m}Tc-MIBI imaging may also be useful as a predictive test or follow-up of response of KS to treatment.

CHAPTER 1

INTRODUCTION

1.1 Background

Acquired immunodeficiency syndrome (AIDS) has reached pandemic proportions with major social and economic impact on society. A global summary by UNAIDS and the World Health Organisation in 2004 reported that 39.4 million people were living with the human immunodeficient virus (HIV). HIV prevalence is highest in South Africa where in 2001 an estimated 4.7 million people were living with HIV/AIDS (UNAIDS, 2004; Cassol *et al*, 2005:324).

Patients infected with the HIV in the developing world, often present very late during the course of the disease, usually with active opportunistic infections (OIs), with tuberculosis (TB) being the most common (Flanigan et al, 2005:321). The interplay between HIV and OIs needs to be better understood so that the impact of OIs on AIDS patients may be addressed. Early diagnosis and treatment of infection and malignancy could improve the quality of life in patients with HIV infection and reduce the high morbidity and mortality associated with AIDS. There is no accurate method to distinguish malignancy from infection in AIDS patients (Abdel-Dayem *et al*,1996: 1662).

Kaposi's sarcoma (KS) is the most common tumour in AIDS patients (Krown, 1999: 580). It has been reported as the initial manifestation of AIDS in about 30% of cases and is the most frequently reported tumour in many African cancer registries (Holkova *et al*, 2001:3848). The increase of KS in the South African population has been ascribed to the epidemic of HIV infection (Hartshorne, 2003: 661). KS commonly presents with skin lesions and may also be widely

disseminated to involve extracutaneous areas such as lymph nodes, larynx, pharynx, bones, lungs, brain, gastrointestinal tract and cause lymphoedema of the extremities and genitals (Krown, 1999:580; Madeddu *et al*, 1978:332).

There is a need for a diagnostic tool to detect the extracutaneous involvement of KS so as to better understand the extent and progression of disease in AIDS. Differentiation of pulmonary infection from malignancy in AIDS patients is necessary since more than 10% of AIDS patients have pulmonary disease and pulmonary KS is the most serious form of KS with a high fatality rate (Holkova *et al*, 2001:3848). At present, not all patients with KS are investigated for pulmonary involvement. Due to the invasive nature of a bronchoscopy, only those patients presenting with acute respiratory symptoms undergo this procedure. Since imaging with ^{99m}Tc-Methoxyisobutylisonitrile (MIBI) is non-invasive it might be the diagnostic tool of choice for detecting pulmonary involvement in AIDS patients.

Imaging using Thallium–201 (^{201}Tl) and Gallium-67 (^{67}Ga) has been reported for the detection of extracutaneous extension of KS (Lee *et al,* 1988:1233) and to distinguish intrathoracic KS from lymphoma and infection (Abdel-Dayem *et al*, 1996:1662). However the difficulty with availability, high cost, increased radiation exposure and unsuitable physical properties of both ^{201}Tl and ^{67}Ga limits the use of these radiopharmaceuticals as compared to Technetium-99m (^{99m}Tc).

^{99m}Tc-MIBI imaging might thus become an important diagnostic tool necessary to help improve the quality of life of AIDS patients.

^{99m}Tc-pertechnetate (^{99m}Tc-O_4^-) may be eluted from the Molybdenum (^{99}Mo) generator, which is commonplace in most nuclear medicine departments. ^{99m}Tc-

O_4^- has the advantages of continuous availability, freedom in patient-scheduling, optimal γ-ray energy for the current γ-camera design, and 6-hour half-life that allows administration of a higher dose, shorter acquisition time, better image quality and smaller radiation dose to the patient (Hassan *et al*, 1989:333 and Fukumoto, 2004: 80).

^{99m}Tc-MIBI can be easily prepared by labelling MIBI with ^{99m}Tc-O_4^- eluted from the ^{99}Mo generator. MIBI is available in a freeze-dried kit with a long shelf life of more than a year. ^{99m}Tc-MIBI is a novel radionuclide-imaging agent that selectively accumulates in cells rich in mitochondria. The mechanism of uptake of ^{99m}Tc-MIBI is proportionally related to blood perfusion. Furthermore, it has the advantages of availability, moderate cost, low radiation exposure and favourable physical properties. ^{99m}Tc-MIBI was initially developed as a cationic lipophilic myocardial perfusion-imaging agent with a similar biological distribution pattern as ^{201}Tl. ^{99m}Tc-MIBI has been successfully used to demonstrate various tumours and may be a simple, non-invasive, economical, and accurate tool to assist diagnosis in AIDS patients.

Hence there is a need for investigation of the suitability of ^{99m}Tc-MIBI imaging as a diagnostic tool to detect the extracutaneous involvement of KS and to differentiate pulmonary infection from malignancy so as to better understand the extent and progression of disease in AIDS patients.

^{99m}Tc-MIBI imaging might thus become an important aid to diagnosis that is necessary to help improve the quality of life of AIDS patients.

1.2 Objective Of Study

The purpose of this study was to evaluate ^{99m}Tc-MIBI imaging, in terms of sensitivity and specificity, for detecting extracutaneous involvement of KS, and for differentiating pulmonary infection from malignancy in AIDS patients before and after treatment.

In order to facilitate analysis, the objective was divided into three sub-problems:

1.2.1 To determine the sensitivity and specificity of ^{99m}Tc-MIBI imaging as a diagnostic tool for detecting the extracutaneous involvement of KS before treatment in AIDS patients.

1.2.2 To determine the sensitivity and specificity of ^{99m}Tc-MIBI imaging as a diagnostic tool for detecting the extracutaneous involvement of KS after treatment in AIDS patients.

1.2.3 To differentiate pulmonary infection from pulmonary malignancy using the results of ^{99m}Tc-MIBI imaging in AIDS patients.

1.3 Hypotheses

1.3.1 It was hypothesized that ^{99m}Tc-MIBI imaging would be both sensitive and specific for the detection of extracutaneous involvement of KS before treatment in AIDS patients.

1.3.2 It was hypothesized that ^{99m}Tc-MIBI imaging would be both sensitive and specific for the detection of extracutaneous involvement of KS after treatment in AIDS patients.

1.3.3 It was hypothesized that ^{99m}Tc-MIBI imaging would be able to differentiate pulmonary infection from pulmonary malignancy in AIDS patients.

CHAPTER 2

LITERATURE REVIEW

2.1 Introduction

In the absence of a safe and efficacious vaccine, HIV has reached pandemic levels. Estimates indicate that HIV worldwide has infected 58 million people with mortality of 23 million as a result of opportunistic infection (Weiss, 2001:2073). According to the global summary compiled jointly by UNAIDS and the World Health Organization in 2004, the total number of people living with HIV in 2004 was 39.4 million of which 2.2 million were children under 15 years of age. The people that were newly infected with HIV in 2004 were 4.9 million and AIDS-deaths in 2004 were 3.1 million (UNAIDS, 2004).

Approximately 25.4 million adults and children living with AIDS are in Sub-Saharan Africa. The infection rate is highest in South Africa. The first recorded case of HIV infection in South Africa was in 1982. In a survey in 2002, the estimated prevalence of HIV infection was 11.4% of the total population (Hartshorne 2003:664). More than 6 million people are presently tested positive for HIV. As cited by Cassol *et al* (2005), it is estimated that some 250 000 South Africans will have died of AIDS by the year 2005 (Cassol *et al*, 2005:324).

AIDS was first recognized in New York and California in 1981 as a combined epidemic of *Pneumocystis carinii* (now *Pneumocystis jirovecii)* pneumonia (PCP) and Kaposi's sarcoma (Krigel & Friedman-Kien, 1985:185). The major clinical manifestations of AIDS can be linked to the cellular immunodeficiency that occurs and include a variety of opportunistic infections (OIs) and malignancies

(Steis & Broder, 1985:304). HIV induces the clinical disease syndrome, AIDS, by causing the progressive depletion of $CD4^+$ T-lymphocytes, the linchpin of the immune system (Engels & Goedert, 2005:407). It has been apparent from the beginning of the HIV epidemic in 1981, that persons infected with HIV have an elevated risk for certain cancers, especially KS.

The understanding of AIDS has been reasonably successful; within six months of the first reported case, the cellular nature of the immune deficiency was known and AIDS was reported as an infectious disease (Weiss, 2001: 2073). HIV was first isolated and reported in 1983 and the cloning of the viral genome and the replication and persistence strategy of the organism followed shortly thereafter. The first clinical trial of an antiretroviral drug was introduced in 1984. By the end of 1985, most developed countries had commenced mass screening of all blood and blood-products. This was followed by the development and testing of these antiretroviral drugs in different combinations on patients who had access to them. Approximately ten years and many different combinations later, these drugs have managed to effectively reduce the mortality rate in infected patients (Weiss, 2001:2073).

The benefits of antiretroviral therapy (ART) have been slow to arrive in the developing world especially in Sub-Saharan Africa, which bears a disproportionate burden of the HIV/AIDS epidemic (Flanigan *et al*, 2005:191). The reasons for this are manifold, the most important being the myth that the benefits of ART cannot be replicated in resource-poor countries. Also implied, is that patients in these countries are too ill from OIs to derive the benefits of ART

(Flanigan *et al*, 2005:191). The issue of access to drugs for AIDS seems to be the latest dimension of the AIDS epidemic. Access to medication for those who have HIV/AIDS can mean the difference between a prolonged fairly healthy life or death. (Nadvi, 2001: 14). Unfortunately the high cost of antiretrovirals has been a prohibitive factor for many patients in the developing world, for example, in South Africa, where the majority of AIDS patients visit public-sector health facilities, the ART program was only implemented in 2004.

Africa in particular has suffered from poverty due to civil war, economic and political instability, and disease and hunger. The AIDS epidemic has compounded these problems as the high morbidity rate creates social problems such as AIDS-orphans, and a decreased healthy working population. The ability to diagnose and treat HIV successfully in Africa has far-reaching implications, both socially and in public health, and hence is an urgent priority as it impacts on life expectancy, HIV transmission rates, orphaned children, productivity and economic stability (Cassol *et al*, 2005:329; Nadvi, 2001: 14).

AIDS is associated with a variety of potentially treatable infections underscoring the need for early detection and treatment of these pathogens. It has been estimated that more than 50% of AIDS patients experience infectious or neoplastic pulmonary manifestations. In 60% of cases PCP is isolated as the only pathogen; in 25% of patients it co-exists with other pulmonary pathogens (Sittler *et al*, 1990:73). This frequent occurrence of OIs is a potential confounding factor in the treatment of HIV in Africa (Cassol *et al*, 2005:324). The complex interplay between HIV and OIs needs to be deciphered so as to understand the

impact of OIs on HIV. This understanding could lead to treatment optimisation especially in Southern Africa where OIs play an important role in both pathogenesis of HIV-1 and patient response to therapy (Cassol *et al*, 2005:330). Co-infection with *Mycobacterium tuberculosis* (HHV-8) and its manifestations in the form of tuberculosis (TB) and KS is common, especially in Sub-Saharan Africa. In the developing world, HIV-infected patients tend to present very late during the course of the disease often with active OIs, for example, TB. This causes activation of the immune system and leads to an increase in the viral load and further impairs HIV-suppression (Flanigan *et al*, 2005:191).

The HIV epidemic has dramatically impacted in all fields of medicine; the clinical and pathological evaluation in the field of gastroenterology has created new challenges for the physician. The symptoms, diagnostic histopathological features and response to therapy for many gastrointestinal diseases are similar in both HIV-positive and HIV-negative patients (Orenstein & Dieterich, 2001:1042).

Tuberculosis, diarrhoea and pneumonia are the major causes of death in AIDS patients (UNAIDS, 2000). KS is the most common malignancy associated with AIDS (Krown, 1999:580). Hartshorne (2003:661) reports that in a survey in South Africa, the prevalence of KS has markedly increased. The increase was ascribed to the HIV epidemic. The size of the HIV/AIDS population is increasing as we embark on the third decade of the HIV epidemic, and the first decade of the availability of ART. Control of the AIDS epidemic must eventually come through preventative medicine. Because of the high mortality and morbidity, prompt

diagnosis and appropriate treatment are necessary. There is no rapid and accurate biochemical or imaging method to distinguish infection and malignancy in patients with AIDS (Abdel-Dayem *et al*, 1996:1662-3).

2.1 Kaposi's Sarcoma (KS)

KS was one of the first opportunistic diseases described in association with AIDS-related diseases. It is also the most common tumour arising in HIV-infected patients and is considered an AIDS-defining illness by the Centre for Disease Control guidelines (Krown, 1999: 580; Dezube, 2000:445). It has emerged from being a rarity to the most important neoplasm in most of sub-Saharan African countries (Weiss & Boschoff 2000:677). In the United States of America (USA), KS is about 20 000 times more common in AIDS patients than in the general population (Dezube *et al*, 2004: 236). Ninety to ninety five percent of KS in the USA occur in HIV-infected men (Haramati & Wong 2000:412). The AIDS pandemic has been accompanied by a KS epidemic.

KS is an unusual tumour of vascular origin that has a proliferative component of spindle cells and endothelial cells as well as an inflammatory and angiogenic component. KS is unusual in its clinical manifestation, course and pathogenesis. The lesions may go into spontaneous remission or spread aggressively to involve subcutaneous tissue, mucosa, lymph nodes and internal organs (Sanders *et al*, 2004: 364).

The pathogenesis of AIDS-related KS is multi-factorial and involves a gamma herpes-virus, KSHV/HHV-8, altered expression and response to cytokines and

the HIV-1 transactivating protein, Tat (Radkov *et al*, 2000: 1121; Mitsuyasu, 2000:175; Dezube *et al*, 2004: 236). HIV-1 Tat is a key progression factor of KS as it enhances all the biological steps of angiogenesis and KS progression (Barillari & Ensoli: 2002:321). The HHV-8 was identified in 1994 as being necessary but not sufficient for the development of KS. There is a 30-50% probability that an HIV-seropositive and HHV-8-seropositive person would develop KS within 10 years of dual seropositivity (Cannon *et al*, 2003: 84). Fauci and Lane (2005:1099) state that in HIV disease, the development of KS is dependent on the interplay of a variety of factors, namely, HIV-1, HHV-8, cytokine secretion and immune activation.

KS was first described by a Hungarian, Moriz Kaposi in 1872 as a 'idiopathiisches multiples Pigmentsarkom der Haut'. The disease that Kaposi described resembled more the KS as seen in AIDS than the classic non-AIDS related KS of older men of Mediterranean and Jewish lineage (Schwartz 2004:146, Steis & Broder 1985:304). The clinicopathological classification of KS has evolved into four major groups, namely; classical type, African type, associated with immunosuppressive therapy type and the type associated with AIDS. The clinical skin appearance is similar in the four groups. In AIDS-KS smaller, earlier subtle lesions are more frequently seen. Lesions range from flat maculae and patches to raised plaques and nodules. Older lesions may become confluent and/or ulcerate. The microscopic appearance of lesions is identical in all four groups. Maculae and patches of early KS consist of a proliferation of irregular, jagged capillary sized dermal vessels. Nodules and plaques have the

characteristic combination of spindle cells and slit-like vessels with red blood cells, siderophages and a round cell infiltrate (Strutton 1991:183-185).

As early as 1988, Roth (1988:149) reported that KS had multiple foci and that a virus was probably associated with the AIDS virus. He also suggested that a well functioning immune system is important in controlling KS. Three decades later, there is still extensive research being performed to treat and control AIDS-KS. Although there have been many important advances in the study of KS, it remains a challenge and an enigma. The characteristics of AIDS-associated KS are a multi-focal widespread distribution that may involve lymph nodes, gastrointestinal tract and visceral organs (Zasshi 1993: 612). KS differs from other neoplasms in that its pattern of dissemination suggests multi-centric origin rather than haematogenous metastatic spread from a single primary site. AIDS patients with KS are usually afflicted with stage 3 or 4 disease (Reichert *et al,* 1985:142).

KS commonly presents as cutaneous lesions. The lesions may appear anywhere on the skin of the face, trunk or extremities. Oropharyngeal lesions are seen in the hard and soft palates, the gingival and buccal mucosa and the tonsils and pharynx (Krigel & Friedman-Kien 1985: 189). KS of the oral cavity affects one third of KS patients. Gastrointestinal involvement is seen in at least 40% of patients on initial diagnosis and in 80% at autopsy in the absence of cutaneous disease. KS of the gastrointestinal tract is often asymptomatic. When symptomatic, endoscopic biopsy is usually necessary because these lesions are often submucosal and not detected on barium studies.

Diagnosis of gastrointestinal or thoracic KS is more difficult (Krown, 1999:581). Fifteen percent (15%) of patients with pulmonary involvement have no evidence of mucocutaneous KS. Diagnosis of pulmonary KS is usually guided by respiratory symptoms, chest radiographs, bronchoscopic or mediastinoscopic evidence (Krown, 1999:581; Dezube *et al*, 2004: 240). Cutaneous KS is often complicated by lymphoedema of the extremities, face and genitals.

Skin lesions can be readily identified and diagnosis confirmed by skin biopsy. Skin and mucus membranes are the typical presentation sites for KS with almost 50% of these having systemic disease with multiple visceral site involvement. Schwartz in 2004 cited the post-mortem findings on 24 cases reported by Lemlich *et al*, in 1987, where 50% of the cohort had lymph node involvement, 50% had gut involvement and 37% had lung involvement. Pulmonary involvement has been reported in more than 10% of all AIDS patients and in up to 25% of patients with cutaneous KS (Holkova *et al*, 2001:3848). Pulmonary KS is the most serious form of KS with high fatality rate and a median survival time of 3-10 months (Holkova *et al,* 2001:3848). In the USA where 90-95% of KS occurs in HIV infected men, at least one third show intrathoracic involvement (Haramati & Wong, 2000:412). Pulmonary KS can be a serious complication of AIDS-KS patients and often mimics *Pneumocystis carinii* infection. Visceral KS, such as respiratory and gastrointestinal symptoms can produce significant organ damage requiring immediate and effective treatment. In the lungs, KS appears as erythematous plaques and nodules in the pleura, along bronchi and interlobular

septae and in the submucosa of the tracheo-bronchial tree (Reichert *et al*, 1985:142).

According to Dezube *et al*, (2004:237), the initial evaluation of patients with AIDS-KS currently involves a clinical examination with endoscopy, occult blood testing for gastrointestinal involvement and chest radiography to screen for pulmonary involvement. Bronchoscopy is reserved for patients that have abnormalities on the chest radiograph or when they have persistent respiratory symptoms (Penney, 1990:108; Lilenbaum & Ratner 1994:142). The time elapsed between the initial negative chest x-ray and re-examination leading to a positive bronchoscopy could considerably delay the diagnosis and treatment of pulmonary involvement. In a study by Haramati & Wong in 2000, the diagnosis of intrathoracic KS involvement on three female patients was only made on autopsy. The delay in diagnosis and hence appropriate treatment could possibly be avoided and the appropriate treatment could be commenced earlier, if a more reliable diagnostic imaging tool was available to adequately detect pulmonary involvement. The ^{99m}Tc-MIBI scan is possibly such a diagnostic tool.

KS lesions are often most likely detected by pathologists on skin biopsies, or on biopsies of the gastrointestinal tract obtained on endoscopy to investigate weight loss, diarrhoea and/or bleeding; or on lung biopsies to rule out haemoptysis or OIs (Reichert *et al*, 1985:147-8).

Early detection of KS is important, since it appears more likely to improve if treated at an early stage. In those patients who present with HHV-8 results that suggest a high risk of KS, in-depth examinations including chest radiology to

detect pulmonary KS should be performed (Cannon, Laney & Pellet, 2003: 84). The prognosis of KS is poor with many patients dying within several months to a few years after onset. Most of these AIDS-KS patients die as a result of severe OIs and not as a direct result of KS (Di Carlo *et al*, 1985:180). Cannon and colleagues in 2003 referred to the article of Dupont *et al,* (2000), stating that if KS and its associated diseases are detected and treated early, the response to ART is improved Cannon *et al,* (2003). Treatment of KS has to be individualized to the needs of the patient. The overall AIDS-related prognosis and aspects of underlying disease such as cytopaenias and OIs will determine the treatment approach selected for AIDS-related KS patients (Mitsuyasu, 2000:176). The comprehensive study by Mocroft *et al* in 2004 concludes that there has been a sizeable decrease in the incidence of KS in the European HIV-1-positive patients since the introduction of highly active antiretroviral therapy in 1994. The treatment of HIV/AIDS is outside the scope of this work and will not be discussed.

Prior to the AIDS epidemic the incidence of KS peaked at around 70 years of age for non-African countries and between 45-50 years of age for East African countries. The incidence of KS was higher in males than in females. However, in a study done by Mwanda *et al*, in 2004 in Kenya on an East African population, the age-group with the highest prevalence of KS was 31-40 years and an identical incidence in men and women was reported. The mean survival time was 104 days; 90% of their cohort succumbed to pulmonary infections and/or visceral involvement of KS.

The extracutaneous spread of KS is common. KS-involvement has been reported to include lymph nodes, larynx, pharynx, bones, lung, brain and the gastro-intestinal tract. The diagnosis of visceral lesions, which may remain clinically silent for a long period is often difficult and is often only evident at autopsy (Madeddu *et al*, 1978:332).

KS and its complications remain a significant challenge to clinicians. Patients with minimal cutaneous disease may benefit from local therapies such as radiation therapy, phototherapy, intralesional chemotherapy. Chemotherapy and/ or treatment with interferon – α has an important role in the systemic treatment of KS that is disseminated to visceral organs, that is, lungs, lymph nodes and gastrointestinal tract (Mitsuyasu, 2000:178). In order for effective treatment to be administered, an easily accessible method for the detection and evaluation of such involvement needs to be available. To date such a diagnostic tool has not been reported in the literature reviewed.

KS often impairs quality of life when it causes disfigurement and leads to social isolation. No simple, non-invasive, and economical method is reported in the literature to determine the extracutaneous extension of KS and response to treatment. Thallium-201 (^{201}Tl)-chloride and gallium-67 (^{67}Ga)-citrate scintigraphy has been used successfully for diagnosing extracutaneous KS (Lee *et al*, 1988:1233; Caceres & Chandeysson 1989:1317). The use of combined ^{99m}Tc-pertechnetate and ^{67}Ga-citrate imaging for the evaluation of KS, was reported by Madeddu *et al,* in 1978. It was later reported that ^{99m}Tc-pertechnetate imaging had a low sensitivity and that it was unable to differentiate between pseudo-KS

and KS. Presant *et al (1990),* cited Woolfenden *et al*, (1987) as reporting that in AIDS patients with PCP, ^{67}Ga was used to image inflammatory lung lesions but did not accumulate in sarcoma lesions (Presant *et al*, 1990:1308). However, ^{201}Tl and ^{67}Ga are expensive and not readily available in South Africa. Limited availability, high cost, increased radiation exposure, and unsuitable physical properties limit the use of ^{201}Tl and ^{67}Ga. Serial scans after ^{67}Ga injection for three consecutive days can also cause difficulty in patient scheduling and hence a delay in diagnosis.

In 2001, a study using ^{111}Indium – DTPA-N-TIMP-2 for the evaluation of KS associated with HIV was undertaken by Kulasegaram *et al*. They concluded that it was unlikely to be of use in KS imaging as the tracer distributed predominantly in the kidneys. No other tissues or KS lesions were identified.

Present *et al* in 1990 report success with imaging KS and lymphoma on two AIDS patients using ^{111}In-labeled liposomes. However the availability, labelling and physical characteristics of ^{111}In-liposomes are problematic.

It is evident from the literature reviewed, that a simple, relatively non-invasive, easily available and economically viable method of evaluation of KS is not available. Improved methods of detection of KS would contribute to better management of AIDS patients with KS. Pulmonary or mediastinal involvement is usually a late manifestation of KS. Chest radiographs may not be of contributory value. Bronchoscopy is necessary for diagnosing endobronchial lesions. Since bronchoscopy may be unreliable in up to 30% of patients with pulmonary KS, open lung biopsy may be required (Krown, 1999:581; Lee *et al,* 1991:409). The

radiographic appearance of PCP is indistinguishable from KS. Prompt detection and treatment reduce patient morbidity and mortality (Cannon *et al,* 2003:85). Sequential ^{201}Tl and ^{67}Ga scans have been used to distinguish KS from lymphoma and infection (Lee *et al* 1991:409, Abdel-Dayem *et al* 1996:1667, Kramer *et al* 1989:671, Reinders Folmer *et al* 1986:313). False-positive Gallium scans have been reported in patients with various OIs (Lee *et al* 1991:409 and Krown 1999:581).

Studies using ^{99m}Tc-MIBI have been reported by Hassan *et al* in 1989 describing uptake in malignant tumours showing a prominent difference between malignant and benign lesions. Aktolun *et al* in 1991 did a study on a single patient with ^{99m}Tc-MIBI and ^{201}Tl to show pulmonary actinomycosis. Kao *et al* in 1993 reported the limited use of ^{99m}Tc-MIBI imaging in the differentiation of single solid lesions in the lungs. In 1998, Richard *et al* suggested further investigation of the use of ^{99m}Tc-MIBI in detecting and monitoring pulmonary involvement in patients with systemic sclerosis. Moustafa *et al* (2003) reported on ^{99m}Tc-MIBI being a reliable imaging tool for the follow-up of bone and soft tissue sarcoma patients to detect recurrence. In a study undertaken by Naddaf *et al* in 1998, where comparison was made between ^{201}Tl and ^{99m}Tc-MIBI brain imaging to differentiate intracranial lymphoma from non-malignant lesions in AIDS patients, ^{99m}Tc-MIBI was found to have a higher sensitivity than ^{201}Tl and hence to be more helpful. They also comment of the preference of ^{99m}Tc-MIBI as compared to ^{201}Tl for imaging intracranial lesions in children because of the better physical characteristics of ^{99m}Tc-MIBI, which allow for higher doses and shorter

acquisition times. The shorter acquisition time is helpful in children who are usually restless.

Although all of these studies used ^{99m}Tc-MIBI imaging, none of the studies were done on patients with AIDS-KS. Some of the studies reviewed, had minimal sample sizes, for example, the study done in 1991 by Aktolun *et al* involved a single patient.

It is evident from the literature reviewed that a reliable, easily available diagnostic tool is required for the detection of extracutaneous and pulmonary involvement of KS.

2.3 Pulmonary Involvement

A number of modalities may be used for the diagnosis of pulmonary manifestation of AIDS. The methods outlined by Sittler *et al* in 1990 include:

2.3.1 Sputum analysis

The evaluation of induced sputum for PCP, although inexpensive and non-invasive, has a sensitivity of only 56%.

2.3.2 Bronchoscopy with Broncheolar Lavage (BAL)

BAL is used to check for the presence of PCP. BAL has to be performed with a transbronchial biopsy done on bronchoscopy that is invasive and uncomfortable for the patient. Bronchoscopy is also indicated where treatment response is poor in cases of PCP diagnosed on sputum analysis. Bronchoscopy with BAL has a high sensitivity for PCP in AIDS patients. BAL may also be used to diagnose

other OIs. Patients with endobronchial lesions often do not undergo biopsy due to the risk of haemorrhage (Haramati & Wong 2000:413).

2.3.3 Fiberoptic Transbronchial Biopsy

Fiberoptic bronchoscopy with transbronchial biopsy (TBB) performed under fluoroscopy is extremely sensitive (98%) for PCP diagnosis. Although BAL and TBB are relatively safe and expedient for many infections in AIDS, their yield in diagnosis of pulmonary KS is low (Sittler *et al*, 1990:76).

2.3.4 Open –Lung Biopsy

Open-lung biopsy although highly sensitive (88%) is not routinely employed in assessing pulmonary involvement in AIDS as the morbidity of an open-lung biopsy is not always an acceptable exchange for the limited therapeutic gain. In studies reported by Sittler *et al,* 1990:85, [Marchevsky *et al* (1985), Mobley *et al* (1985) and Wallace & Hannah (1982)], at autopsy, the number of AIDS patients who died with potentially treatable, but undiagnosed disease ranged from 50 – 68%. Many of these infections are potentially curable; hence their early detection is essential in the management of AIDS patients.

PCP is recognized as a cause of pneumonia in immunosuppressed patients. PCP is the most life-threatening infection in AIDS patients. It presents a special diagnostic problem as symptoms are masked by fever, malaise, cough and dyspnoea and the chest x-ray is usually normal in early PCP. The usual pattern of Gallium uptake for patients with PCP is diffuse and bilateral. However in a study done by Charron *et al* in 1988, they found focal uptake in the chest, which

is usually characteristic of OIs other than PCP. They concluded that the focal uptake seen in their patients was possibly early presentation of PCP.

Mycobacterial infections are common, but treatable infections in AIDS patients, provided they are diagnosed early. Diagnosis is often delayed due to the atypical clinical and radiographic presentations. This is detrimental to the patient and to public health. Hence Lee *et al* in 1994, reported that ^{201}Tl-positive and ^{67}Ga-negative pattern of uptake in AIDS-KS patients was indicative of mycobacterial infection.

Abdel-Dayem (1989, 1994, 1996, 2000) has been involved in many different nuclear medicine imaging studies of the pulmonary area in AIDS patients. In 1994 Abdel-Dayem *et al*, did a study on five AIDS patients with PCP that showed diffuse bilateral lung uptake on sequential thallium and gallium scans. They concluded that in AIDS-KS patients, ^{201}Tl-uptake is usually focal and not diffuse unless there is some other pathology, for example, PCP, present. In the study done by Abdel-Dayem *et al* in 1996, on sequential ^{201}Tl and ^{67}Ga scans of the chest in AIDS patients, they concluded that a ^{201}Tl-positive and ^{67}Ga-negative pattern of uptake in AIDS patients showed a high specificity (89%) for KS, however the sensitivity decreased to 37% in the presence of opportunistic infection.

The need for differentiating the different OIs or malignant involvement of the lungs in AIDS – KS patients is necessary so as to facilitate appropriate treatment. Abdel-Dayem *et al* (1996) identified several problems on a retrospective evaluation of the studies that had been done thus far involving sequential ^{201}Tl

and ^{67}Ga scans of the chest to differentiate KS from OIs and malignancy in AIDS patients. The problems they listed were:

- AIDS patients usually have more than one pathology at any given time (more than one opportunistic infection with KS and/or malignant lymphoma of the lungs)
- Difficulty in confirming diagnosis in AIDS patients due to low sensitivity of laboratory tests and the time needed for sputum culture in infections such as TB.
- Invasive procedures such as bronchoscopic broncheolar lavage (BAL) or biopsy that were needed to confirm diagnosis were exhausting to the patient.
- Chest imaging was non-specific and not specific especially in the early phase of infections such as PCP.

2.4 Radiopharmaceuticals

^{99m}Tc-MIBI was initially developed as a myocardial perfusion agent with a similar biological distribution pattern as ^{201}Tl. Although ^{99m}Tc-MIBI is a lipophilic imaging agent empirically designed for the non-invasive evaluation of coronary artery disease by external scintigraphy, over the last decade it has been widely used for tumour imaging (Naddaf *et al*, 1998:19).

^{99m}Tc is available and produced daily in most nuclear medicine departments from a Molybdenum-99 (^{99}Mo) generator. The radiopharmaceutical, MIBI, is available in a freeze-dried kit form and has a long shelf-life of usually over a year. ^{99m}Tc-

MIBI can be easily prepared by labelling MIBI with ^{99m}Tc-pertechnetate ($^{99m}TcO_4^-$) eluted from the ^{99}Mo generator by most nuclear medicine technologists / radiographers or radiopharmacists. ^{99m}Tc-MIBI has the advantages of continuous availability for use, freedom in patient-scheduling, smaller radiation dose to the patient and ideal physical characteristics for use with the current gamma-camera design (Fukumoto, 2004:87; Hassan *et al*, 1989:333).

^{99m}Tc effectively emits a single gamma photon with a photon energy of 140 keV compared to ^{67}Ga with three energy peaks of 92, 182, and 364 keV, and ^{201}Tl with x-ray photon energy of 80 keV and two gamma energy peaks of 135 and 167 keV (Theobald 1994:103). The current design of gamma camera produces optimal images for 140 keV, hence the photon energy of ^{99m}Tc is ideal. Image quality is inferior for ^{67}Ga and ^{201}Tl.

The shorter physical half-life of ^{99m}Tc of 6 hours is a definite advantage in terms of minimal radiation exposure to the patient as compared to the half life of ^{67}Ga of 78 hours and that of ^{201}Tl of 72 hours which result in an increased radiation burden to the patients (Fukumoto, 2004:80; Theobald 1994:103).

^{99m}Tc-MIBI has several advantages compared to ^{201}Tl-chloride. $^{99m}TcO_4^-$ may be available to be labelled with MIBI for use in patients at any time in the nuclear medicine laboratory as opposed to ^{201}Tl and/or ^{67}Ga that has to be specifically ordered per patient dose for delivery and administration on certain set days. The optimal gamma-ray energy and 6-hour half-life of $^{99m}TcO_4^-$ allow for the administration of a higher dose so as to get better resolution. In addition ^{99m}Tc-

MIBI has been proposed as a tracer to measure P-glycoprotein (P-gp) function that has an important role in tumour multi drug resistant to chemotherapy.

The mechanism of uptake of ^{99m}Tc-MIBI is proportionally related to the transmembrane potential of tumour cells as well as blood flow and capillary permeability indirectly responsible for tumour uptake. ^{99m}Tc-MIBI, with properties of cationic charge and lipophilicity, crosses the capillary endothelium, diffuses through the interstitial space, transports across the tumour cell membrane, localizes in mitochondria, and/or attaches to a low molecular weight protein in the lysosomes (Shih *et al*, 1998:507). In vivo ^{99m}Tc-MIBI selectively accumulates in cells rich in mitochondria (Richard *et al*, 1998:19). The uptake of ^{99m}Tc-MIBI in tumour cells could be linked to the presence of mitochondria as it accumulates more intensely in malignant tumour cells than in their surrounding epithelial or connective tissue (Fukumoto, 2004:82). ^{99m}Tc-MIBI is taken up by normal tissues that are rich in mitochondria, such as cardiac muscle and liver tissue and by tumours (Richard *et al*, 1998:19). Accumulation of ^{99m}Tc-MIBI in tumour tissue depends on mitochondrial and plasma protein potentials, intracellular mitochondrial density, expression of P-gp, and blood flow (Furata *et al*, 2003: 982). Hence ^{99m}Tc-MIBI has been successfully used to demonstrate tumours of the brain, nasopharynx, thyroid, parathyroid, lung, breast, kidney, and lymphoma. Based on this knowledge it might be possible to demonstrate extracutaneous and pulmonary KS using ^{99m}Tc-MIBI.

The uptake of ^{99m}Tc-MIBI in tumours is affected by a number of variables. The washout of ^{99m}Tc-MIBI from tumour cells is related to the multi-drug resistant

energy dependent P-gp pump system. Tumour cells with a higher concentration of this transmembrane protein demonstrate a faster clearance rate and hence less tracer uptake. When compared to ^{201}Tl, the lower tracer uptake is not problematic as, the half-life of ^{99m}Tc permits the use of a higher dose that will increase image count rate and give better resolution.

The article by Fukumoto (2004) reports on various other studies done where tumour imaging with ^{99m}Tc-MIBI, validated as a P-gp transport substrate, a predictive test of tumour response to anticancer agents.

Richard *et al*, in 1998, found that uptake of ^{99m}Tc-MIBI was abnormally elevated in lungs of systemic sclerosis patients with pulmonary involvement, where pulmonary involvement was confirmed on clinical, laboratory, and/or radiological evidence. They suggested that a larger study be done on systemic sclerosis patients to determine whether ^{99m}Tc-MIBI-scintigraphy can detect pulmonary involvement at a pre-clinical stage.

Thallium-201 (^{201}Tl) has been successfully used for tumour imaging. ^{201}Tl accumulates mainly in viable tumour tissue with lesser uptake in inflammatory connective tissue and negligible uptake in necrotic tissue. There seems to be a high degree of positive correlation between tumour vascularisation and uptake of ^{201}Tl. Although ^{201}Tl is recognized as a tumour-imaging agent, some benign pathologies also demonstrate uptake. Only delayed ^{201}Tl-imaging may be helpful in differentiating tumour from infection, as tumours will retain uptake and uptake will wash-out from benign or inflammatory lesions. The sensitivity and specificity

of ^{201}Tl for the detection of lung carcinoma has been reported to be 71-100% and 30-100% respectively (Kao *et al*, 1997:1015).

In a letter to the editor, Abdel-Dayem (1994), cautions on over-reading of pulmonary ^{201}Tl scans as uptake in the right ventricle may be misinterpreted as positive pulmonary uptake. Lee et al in 1989, reported on a single patient where ^{201}Tl scintigraphy was able to demonstrate the extent of tumour in a KS patient and they recommended the potential use of ^{201}Tl to determine the presence and extent of cutaneous and extracutaneous AIDS-KS.

^{99m}Tc-MIBI has superior imaging characteristics when compared to ^{201}Tl, and this has limited its (^{201}Tl) clinical applications as a tumour-seeking agent (Moustafa *et al,* 2003:51). Imaging with ^{99m}Tc-MIBI is possible shortly after the administration of the dose, since maximum tumour uptake is reached within one minute (Hassan *et al*, 1989: 333).

Fukomoto in 2004 reports that in a study done by Ando *et al* in 1987 on the bio-distribution of ^{201}Tl, and that ^{201}Tl was more specific than ^{67}Ga in differentiating tumours from benign and inflammatory lesions. It was also noted that the bio-distribution of ^{201}Tl and ^{67}Ga was different and that their mechanisms of uptake were different. However promising ^{201}Tl imaging might seem in differentiating tumour from infection, ^{201}Tl has unsuitable physical characteristics; the long half-life of 72 hours is restrictive in terms of dose administration that then impacts on the resolution of the final image.

^{67}Ga imaging has played a valuable part in evaluation of HIV. The use of ^{67}Ga as a tumour-imaging agent is limited due to its lack of tumour-specificity and inability

to detect tumours smaller than 2 cm in diameter (Kao *et al,* 1997:1015). Getz and Bekerman (1994) report the role of ^{67}Ga in the detection of pulmonary and extrapulmonary KS to be extremely limited. KS lesions in ^{67}Ga scintigraphy are usually negative whereas increased uptake is seen on ^{201}Tl imaging (Del Val Gomez *et al*, 1994:467). However, Krishna & Chitkara (2003) report on a case where both ^{210}Tl and ^{67}Ga scans were negative on osseous KS lesions that were confirmed on CT guided-biopsy. They questioned the reliability of sequential ^{210}Tl and ^{67}Ga scintigraphy in osseous KS evaluation.

In lymphomas, both ^{67}Ga and ^{201}Tl images are positive, but a similar pattern is noted in KS patients with TB. Hence the combination of ^{67}Ga and ^{201}Tl scintigraphy is not a reliable option in differentiating malignancy from infection in KS patients.

Although ^{67}Ga has a role in establishing a diagnosis, assessing location and extent of disease, differentiating active disease from necrotic tissue and determining recurrence or response to therapy in certain lung diseases, the use of ^{67}Ga is hampered by its lack of specificity, typical 1-3 day delay between injection and final imaging and sub-optimal imaging characteristics (Schuster & Alazraki, 2002:193). Turoglu *et al* in 1998, reported a new trend in the ^{67}Ga-pattern in PCP-infected patients, where the intensity of uptake in the lungs was mild and the diagnosis could be missed. There still exists a need for a diagnostic imaging tool for lung disease.

A study in 1982 performed by Gunnoe and Kalivas reported that ^{99m}Tc-O_4^- imaging was positive in both KS and pseudo-KS and hence not helpful to

differentiate one from the other. Rosen, Martin & Stern in 1979, reported that ^{99m}Tc-O_4^- imaging was not useful in differentiating KS from pseudo-KS. Another study by Williams *et al* in 1985 employed ^{99m}Tc-O_4^- for imaging KS. They reported that using ^{99m}Tc-O_4^- imaging was relatively insensitive for KS. Witte *et al* in 1990, performed lymphoscintigraphy using ^{99m}Tc-Human Serum Albumin (HSA) on a cohort of AIDS-KS patients to evaluate the nature and extent of lymphatic involvement. This study confirmed the lymphatic involvement in AIDS associated KS patients.

^{99m}Tc-Tetrafosmin (^{99m}Tc-Tf) has been successfully used as a myocardial-imaging agent. Following the usefulness of ^{99m}Tc-MIBI and ^{201}Tl as tumour-imaging agents, the potential of ^{99m}Tc-Tf has been proposed as a possible tumour agent (Fukumoto, 2004:80). Although ^{99m}Tc-Tf-uptake seems similar to ^{99m}Tc-MIBI-uptake, the mechanism of uptake is not yet understood. It seems as if only a small fraction of ^{99m}Tc-Tf accumulates in the mitochondria whereas ^{99m}Tc-MIBI-uptake is intra-mitochondrial. Kao *et al* (1997:1017) comment on studies that report on the retention of ^{99m}Tc-MIBI in cells being dependent on the activity of the 170kDa P-gp, coded by multidrug resistance gene, which functions as an ATP-dependent efflux pump for cytotoxic substances. ^{99m}Tc-MIBI is reported to be a ligand for multi drug resistance (Kao *et al*, 1997:1017). Positive tumour uptake of ^{99m}Tc-MIBI is considered a low expression pf P-gp. ^{99m}Tc-Tf is a ligand for the same P-gp (Kao *et al*, 1997:1017). Hence ^{99m}Tc-Tf and ^{99m}Tc-MIBI are similar in *in-vivo* use being a substrate for P-glycoprotein (P-gp), a membrane transport responsible for multidrug resistance.

^{99m}Tc-Tf has been reported as having a lower uptake than ^{99m}Tc-MIBI in breast tumours (Kao *et al*, 1997: 1018). It also shows higher background activity than ^{99m}Tc-MIBI in thyroid cancer. Hence ^{99m}Tc-Tf was considered not a good alternative to ^{99m}Tc-MIBI and of little or no clinical value in the detection of lung cancers (Kao *et al*, 1997: 1018). In a letter to the editor, Ohtake (1998), comments on the decreased sensitivity of ^{99m}Tc-Tf in detecting small cell carcinomas especially in bronchoalveolar adenocarcinoma. He also comments that in the case of a large mass with poor perfusion of ^{99m}Tc-Tf, the lesions will only be partially visualized or not visualized.

The usefulness of ^{99m}Tc-Tf SPECT imaging for the detection of intrathoracic malignant lesions was reported (Spanu *et al* 2003a: 639). However there is no indication as to the HIV status of these patients. In another study by Spanu *et al*, 2003b: 295, to show the usefulness of ^{99m}Tc-Tf in different variants of KS, on 27 KS patients of which only 5 were AIDS-KS, SPECT imaging was found to be useful in both detection and staging of KS lesions.

In 2003, Song *et al* concluded that the slow tumour clearance of ^{99m}Tc-MIBI can predict a good response to chemotherapy in non-Hodgkin's lymphoma patients. Kawata *et al* in 2004, reported that ^{99m}Tc-MIBI imaging was useful to suggest the response to chemotherapy in advanced gastric cancer patients.

In a study by Wu *et al* in 2003, it was concluded that ^{99m}Tc-Tf SPECT imaging was a useful additional tool to detect metastases in papillary thyroid cancer but small lymph nodes and miliary lung metastases may be missed.

Kao *et al* (1997:1017) cited studies done by Biggie *et al* in 1991, where radiolabelled monoclonal antibodies (MAbs) have been reported as being potentially of value for cancer diagnosis. However due to the lack of specificity of these agents, their use has been limited. In a study done by Kulasegaram *et al* in 2001 on the *in-vivo* evaluation of ^{111}In-DTPA-*N*-TIMP-2 in HIV-KS, they concluded that ^{111}In-DTPA-*N*-TIMP-2 could be used as an imaging tracer, but that it was unlikely to be of use in KS imaging due to its lack of uptake by KS lesions.

Positron emission tomography (PET) was introduced into oncology as early as 1994. However due to the practical problems such as high cost and lack of availability associated with Flurodeoxyglucose_PET (FDG-PET), its use is limited (Henze *et al* 2002: 325; Kao *et al* 1997:1015). Furuta *et al* (2003) report that ^{99m}Tc-MIBI imaging is possibly superior to FDG-PET for the detection of tumour recurrence early after radiotherapy. They suggest that a comparative study between ^{99m}Tc-MIBI and FDG-PET for tumour recurrence be carried out on a larger cohort.

Brain SPECT imaging with ^{201}Tl has shown an accuracy similar to that of ^{18}F-FDG-PET for imaging recurrent glioma, however for primary brain tumours ^{99m}Tc-MIBI brain SPECT has demonstrated better sensitivity and specificity than ^{201}Tl-studies (Beauchesne *et al*, 2004: 409-412). A comparative study between ^{18}F-FDG-PET and ^{99m}Tc-MIBI in the assessment of patients with multiple myeloma showed that ^{99m}Tc-MIBI scans detected more disease sites than ^{18}F-FDG-PET images (Mileshkin *et al*, 2004:36).

The cost, as calculated from the vendor's quotations in Durban, South Africa, of approximately R200 per patient for ^{99m}Tc-MIBI compares favourably to R850 for ^{67}Ga and R1100 for ^{201}Tl. By reconstituting one vial of MIBI to yield multiple doses for tumour and cardiac imaging on the same day, the cost of ^{99m}Tc-MIBI can be further reduced. There has been a continuous effort to find a tracer that could be labelled with ^{99m}Tc for imaging of lung carcinoma because of the ready availability and attractive nuclear properties of ^{99m}Tc for planar and SPECT imaging. ^{99m}Tc-MIBI has been considered as having the largest potential to fulfil this need (Kao *et al*, 1997:1017). Nuclear medicine imaging has the advantage that it does not only provide morphological information, but also gives important indications of physiology and biology of cancer.

It is important to detect extracutaneous involvement and to differentiate pulmonary infection from malignancy in AIDS-KS patients with regard to treatment and prognosis. It is evident from the discussion above that ^{99m}Tc-MIBI has several distinct advantages over the other radiopharmaceuticals mentioned. ^{99m}Tc-MIBI could prove to be the simple, easily available, affordable radiopharmaceutical with well-suited physical characteristics to provide a non-invasive test to detect extracutaneous KS and to distinguish infection from malignancy in AIDS patients with pulmonary disease. It may also be useful as an indicator of tumour drug resistance, and therefore as a predictor of treatment responsiveness, because of its role as a marker for multi drug resistant protein.

CHAPTER 3

RESEARCH METHOD AND DESIGN

3.1 Study Design

This study was a prospective, non-randomised controlled study to determine the sensitivity and specificity of ^{99m}Tc-MIBI imaging for detecting extracutaneous involvement of Kaposi's Sarcoma (KS) and differentiating pulmonary infection from malignancy in AIDS patients.

3.2 Sampling

Permission to conduct the study was obtained from the Durban Institute of Technology ethics committee (Appendix 1). Informed consent (Appendix 2) was obtained from each patient.

Control Group

Nineteen clinically diagnosed AIDS patients referred from the Respiratory Clinic at Inkosi Albert Luthuli Central Hospital (IALCH), with confirmed pulmonary pathology on bronchoscopy were imaged using the protocol described below. The investigator was blinded to the bronchoscopic findings. The results were quantified. The ^{99m}Tc-MIBI imaging results were compared to the bronchoscopy findings. This group of patients formed the baseline/control group of the study.

Sample Group

Using a non-randomised convenience sampling technique, sixty six AIDS patients with biopsy-proven KS referred by the Dermatology Department at King Edward VIII Hospital, were studied to detect extracutaneous KS and possible differentiation of pulmonary infection from malignancy. Participants were imaged

using the protocol described below. A bronchoscopy was performed by the Respiratory Clinic prior to treatment, usually on the same day as the ^{99m}Tc-MIBI imaging.

3.3 Inclusion Criteria

Control Group

i) The selected clinically diagnosed AIDS patient had to have a confirmed pulmonary pathology on bronchoscopy.

ii) The selected patients had to be referred by clinicians from the Respiratory clinic.

iii) The participant had to have been counselled for AIDS.

Sample Group

i) The participant had to have tested positive for HIV/AIDS and KS.

ii) The patients were referred by clinicians from the Dermatology department.

iii) The participant had to have been counselled for AIDS.

3.4 Exclusion Criteria

Control Group and Sample Group

i) Patients who were not HIV positive or who refused to be tested to establish their HIV-status were excluded

ii) Patients who had commenced treatment for HIV and/or KS were excluded.

ii) Due to the use of radioactivity, female patients who were pregnant, lactating or of child-bearing potential, who were not on any contraception were excluded.

The proposal was approved with a sample size of 15 patients in the control group and 75 patients in the screening sample group. Due to the changes implemented in early 2005 by the Department of Health in terms of the availability of antiretroviral medication, the recruitment of patients that satisfied the inclusion/exclusion criteria was severely hindered as most of the patients had already commenced treatment before they could be considered for inclusion in the sample (Madlala, 2005:4). Approval for a reduction in sample-size from 75 to 50 patients for the sample group was obtained from the Centre for Research Management and Development, Durban Institute of Technology (Appendix 3).

Of the 66 patients referred for the sample group, 4 patients had post treatment follow-up studies performed. Twenty-one (21) patients demised prior to follow-up, 11 did not have any treatment, 11 patients were not contactable telephonically or by mail, 17 commenced anti-retroviral treatment, 4 refused bronchoscopy and 2 refused imaging.

It was assumed that those patients, who were given oral medication as part of their treatment regimen, took their medication as directed. The treatment regimen refers to the treatment that the referring doctor thought to be optimal for the patient.

3.5 Method

The age and sex of each patient was recorded. Where available, results of laboratory assessments including CD4 counts and clinical assessments were noted.

So as to add further value to the study, single photon emission tomography (SPECT) imaging of the chest, although not required on the initial protocol, was performed. A comparison of the planar and SPECT images was made. This did not increase the time the patient spent in the Nuclear Medicine Department as the SPECT study was performed during the period between the whole body scan at 20-minutes post injection and the 60-minute chest planar image. However, if the patient was unable to lie still or was not co-operative for the acquisition of the SPECT study, the study was not performed. Thirteen and fifty-four participants from the control and sample cohorts respectively had SPECT imaging.

3.5.1. Imaging Protocol

The patient was given an intravenous injection of 740MBq (20 mCi) ^{99m}Tc- MIBI (Cardiolite, Bristol-Myers Squibb**,** New York, USA). Anterior and posterior planar scans of the chest were acquired at 10 and 60 minutes post injection with the patient in the supine position using a rectangular, large-field-of-view, dual-head gamma camera (Siemens E-Cam) using the low-energy, high-resolution, parallel-hole collimators. A 10% window centred on the 140 keV photopeak, a 256 x 256 matrix, and 750k counts per image were used for the acquisition.

Anterior and posterior whole body scans were obtained at 20 minutes using a scan speed of 12 cm/min. SPECT images were acquired at 40 minutes over 360° using a 64 x 64 matrix, 6° angular step, and acquisition time of 30 s/projection. The body contouring system was enabled so as to ensure a minimum distance between the patient and the collimator during rotation.

3.5.2. Image Processing/Analysis

1. Using the software on the Siemens e-cam system, regions of interest (ROIs) were drawn in the areas showing maximal uptake in the lung, on the ipsilateral arm muscle, and in the myocardium on the anterior and posterior planar images.

2. The number of pixels, average count/pixel and area of ROIs in the lung, arm muscle and myocardium were recorded.

3. The geometric mean (GM) of the average count/pixel of the ROIs on the anterior and posterior images was used to calculate the lung/myocardium and lung/arm muscle ratios at 10 minutes and 1 hour using:

$GM = (\text{Anterior count/pixel} \times \text{Posterior count/ pixel})^{1/2}$

4. The lung retention ratio (LRR) was determined by the equation:

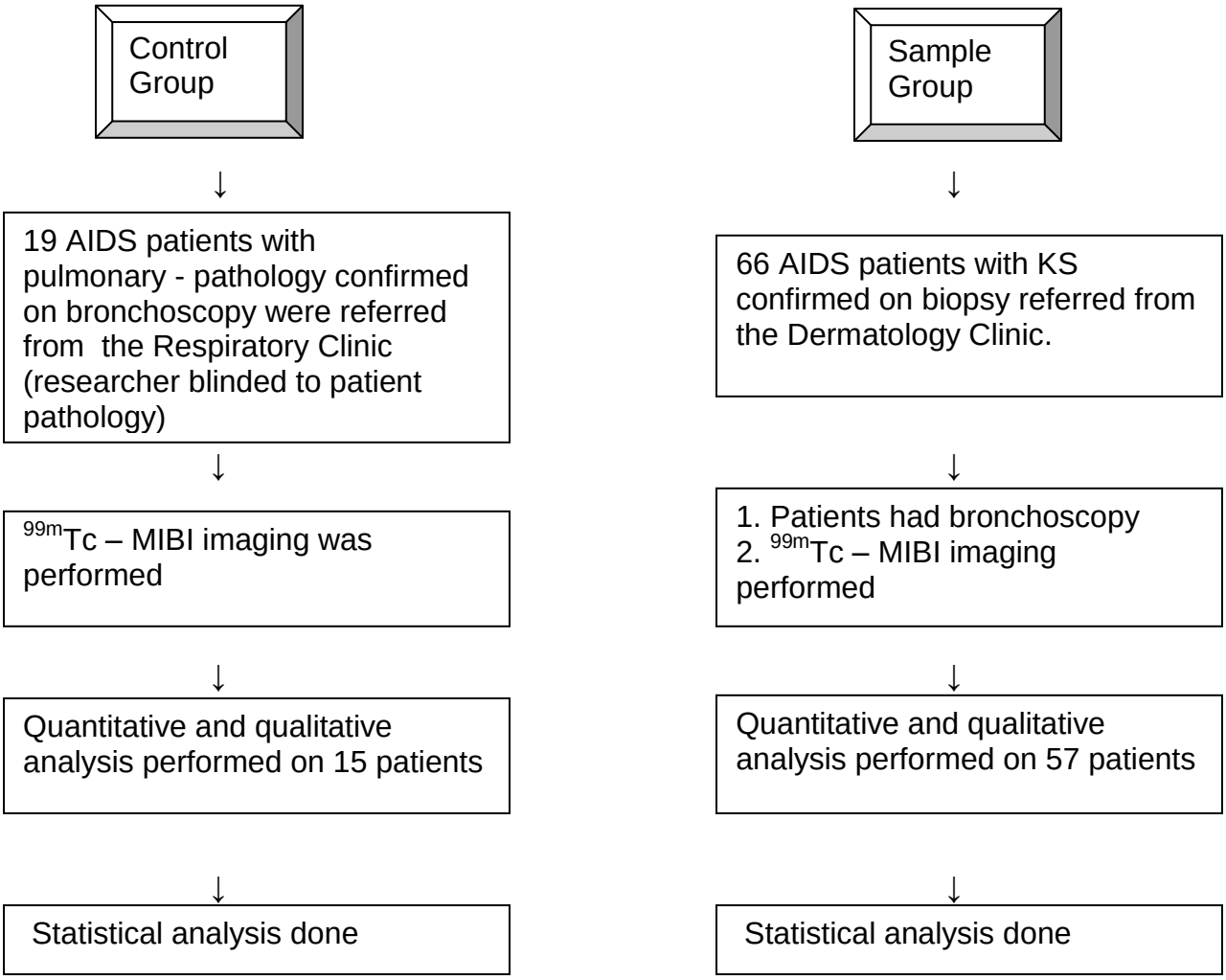
LRR = GM at 60 minutes/ GM at 10 minutes

5. The SPECT images were processed using a Gaussian filter (10 mm full-width-half-maximum) and no attenuation correction was applied to obtain axial, coronal and sagittal images. The lung/myocardium, lesion/lung and lesion/myocardium uptake ratios were determined on the axial SPECT images using average count/pixel of the ROIs

6. A nuclear medicine physician and a medical doctor/ medical physicist with expertise in interpreting nuclear medicine studies reviewed the images simultaneously without knowledge of the clinical findings and results of other investigations.

7. Lung uptake intensity greater than that of arm muscle was interpreted as 'Increased'. The pattern of lung uptake was categorized as no abnormal uptake, diffuse homogeneous uptake, diffuse heterogeneous uptake (if greater than 50% of the total lung area showed areas of greater concentration and areas of relative sparing), or local pulmonary accumulation. Uptake in the axillary, hilar, mediastinal or cervical lymph nodes, pleural or pericardial effusion, or ascites was also recorded.

3.6 Summary Of Research Design



3.7 Statistical Analysis

In order to meet the objectives of the study the results obtained were analysed in separate groups, namely, chest and cutaneous.

3.7.1 Chest Data Analysis

Analysis of planar (10 minute and 60 minute images) and SPECT images of the chest was performed so as to differentiate pulmonary infection from malignancy using the results of ^{99m}Tc-MIBI imaging in AIDS patients. The final patient diagnosis was made by histological, microbiological or bronchoscopic findings. Using the final diagnosis as the gold standard, sensitivity, specificity, and accuracy of the visual interpretation and quantitative analysis methods was determined.

All the patients studied were HIV positive. The data obtained from the chest images was divided into 4 groups as shown in figure 1:

a. KS patients with KS lung involvement (n=36)

b. KS patients with no KS lung involvement (n=11)

c. KS patients with other lung pathology (n=10)

d. Non-KS patients with other lung pathology (control group, n=15)

Although 19 patients were referred for the control group and 66 patients were referred for the sample group, some images were sub-optimal and were not suitable for inclusion in the study.

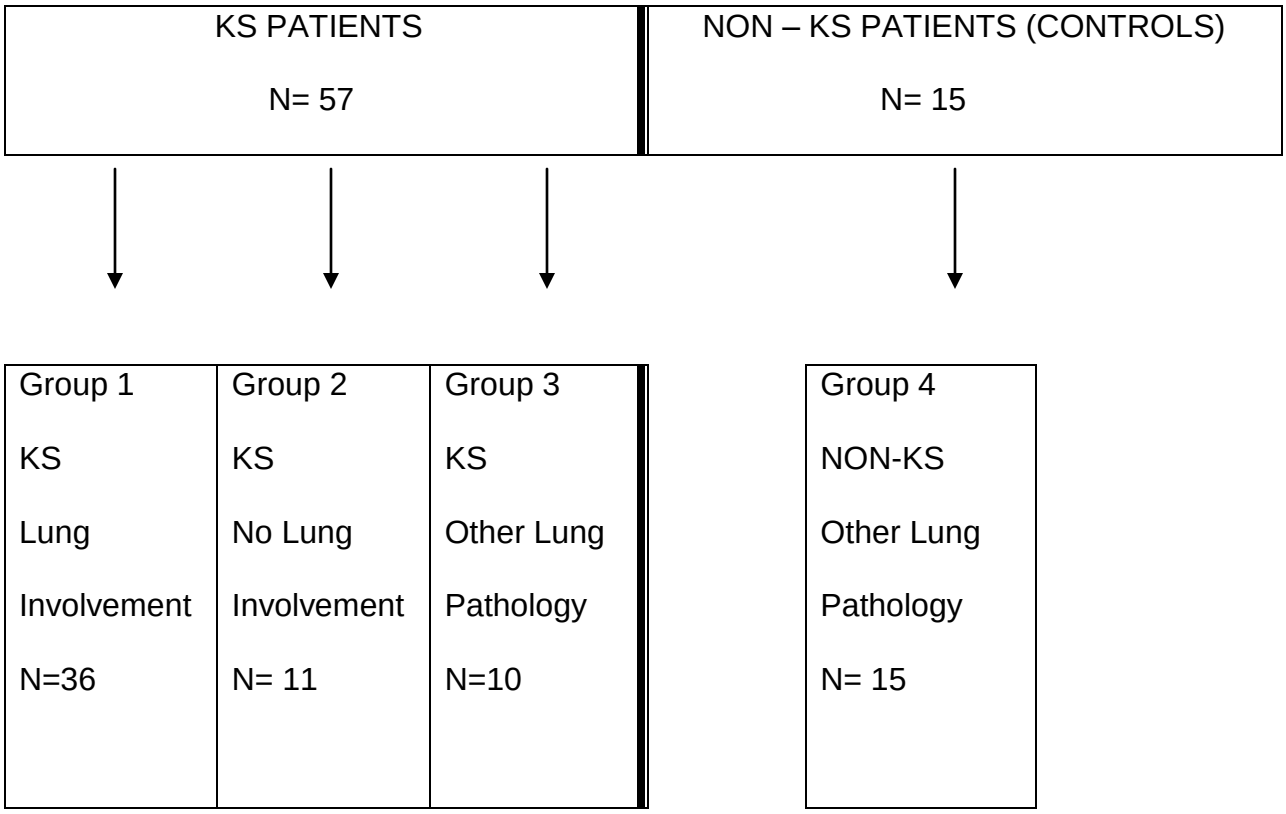


Figure 1: Diagram of chest study sample showing 4 subgroups

Following the diagram in Figure 1 above, comparisons were made between the different groups listed below and the results were analysed to include value power of the statistical tests. The groups analysed were:

i) HIV positive KS patients: lung involvement vs. no lung involvement (Groups 1 and 3 vs. 2)

ii) HIV positive KS patients: KS lung involvement vs. other pathology (Group 1 vs. Group 3)

iii) HIV positive KS patients: KS in lungs vs. normal lungs (Group 1 vs. Group 2)

iv) HIV positive patients: KS in the lung vs. other pathology (Group 1 vs. Groups 3 and 4)

v) HIV positive patients: lung involvement vs. normal lungs (Groups 1, 3 and 4 vs. Group 2)

vi) HIV positive patients: KS in lung vs. all others (Group 1 vs. Groups 2, 3 and 4)

vii) HIV positive patients: KS vs. non-KS (Group 1, 2, and 3 vs. 4)

viii) HIV positive patients: KS in the lungs vs. non KS other lung pathology (Group 1 vs. group 4)

3.7.2. Cutaneous Data Analysis

Analysis of planar whole body images was performed so as to determine the efficacy of ^{99m}Tc- MIBI imaging as a diagnostic tool for detecting the cutaneous involvement by KS in AIDS patients.

Because of normal radioactivity in the salivary glands, pharynx, myocardium, hepatobiliary, gastrointestinal, and urinary tracts that obscures the face and trunk, assessment of cutaneous lesions on whole body planar imaging was confined to the extremities. Soft tissue swelling and any foci of increased ^{99m}Tc-MIBI-uptake in the extremities and lymph nodes were recorded. Comparison was made with clinical assessment by dermatologists for the following:

a. Lymphoedema

b. Lymph nodes

c. Extent of involvement

d. Skin description

SPSS version 11.5 (SPSS Inc. Chicago, Ill, USA) was used for data analysis. Relative errors and ratios were analysed descriptively for the available data in the sample using means, medians and inter-quartile ranges. Ratios were compared between the groups using non-parametric Mann-Whitney tests. Bonferroni adjustment was used to account for the increased probability of a type I error due to multiple tests and sub-setting of the data. Since 8 subsets were examined, the alpha level used to assess statistical significance was 0.00625 (ie.0.05 /8). ROC analysis was used to assess the cut points of the ratios that would optimise sensitivity and specificity to predict certain outcomes. This was also done on specific subsets of the data. LLR (local linear regression) smoothing was used to create a smoothed fit line for the ROC curve. Sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) and their 95% confidence intervals were calculated using Epicalc 2000 version 1.02 (Gillman & Myatt, 1998). Kappa statistics for agreement were calculated using Graphpad quickcalcs software freely available on the Internet at the site:http://graphpad.com/quickcalcs/kappa1.cfm

CHAPTER 4

RESULTS

The results are reported in two data sets, namely, chest analysis (Section 4.1) and cutaneous analysis (Section 4.2).

4.1 Chest Analysis Results

4.1.1 Ratios – Overall

The ratios of the geometric mean (GM) for planar and SPECT imaging for the different data sets were analysed descriptively using means and medians.

The lung/myocardium uptake ratios were also determined on the axial SPECT images using average count/pixel of the ROIs. Because the skewness of the data on descriptive statistics showed that the distribution was not a normal distribution, the ratios were compared using the non-parametric Mann-Whitney U method.

4.1.1 (i) Statistical Methods Used in the Analysis

The Wilcoxon Mann-Whitney Test is one of the most powerful of the non-parametric tests for comparing two populations. It is used to test the null hypothesis that two populations have identical distribution functions against the alternative hypothesis that the two distribution functions differ only with respect to location. The Wilcoxon Mann-Whitney Test does not require the assumption that the differences between the two samples are normally distributed (Lane, 2005).

In statistical terms correlation is used to denote association between two quantitative variables. The degree of association is measured by a correlation coefficient. The Spearman rank correlation coefficient is an example of a correlation coefficient. It may be used to check the relationship between two variables when the relationship is non-linear as is assumed with the data being compared in this study (Lane, 2005; bmj.bmjjournals, 2004). In this study, the Spearman's correlation was done to examine the relationship (if any) between CD4 counts and the ratios. Since the ratios were not normally distributed a rank test, which is distribution free such as the Spearman's test, was thought to be appropriate to examine correlation in such data.

Analysis of variance (ANOVA) test is used to test hypotheses about differences between two or more means. ANOVA may be used to test differences among several means for significance without increasing the Type 1 error rate (Lane, 2005). In this study, since age was normally distributed and there were more than 2 groups, the ANOVA test was used to compare the mean age between the different groups.

Table 1: Descriptive statistics for planar ratios - overall

	Ratio GM 10” lung / 10” myocardium	Ratio GM 10” lung / 10” muscle	Ratio GM 60” lung / 60” myocardium	Ratio GM 60” lung / 60” muscle	lung retention ratio GM 60” lung / 10” lung
N	72	72	72	72	72
Mean	0.57	3.86	1.14	2.95	0.95
Median	0.58	3.47	1.08	2.59	0.86
Std. Dev.	0.12	2.04	0.52	1.42	0.32
Minimum	0.27	1.05	0.39	1.02	0.61
Maximum	0.91	10.28	3.28	9.70	2.25

Table 2: Descriptive statistics for SPECT ratios - overall

	SPECT Ratio Lung/Myocardium	SPECT Ratio Lesion/Lung	SPECT Ratio Lesion/Myocardium
N	67	16	16
Mean	0.46	1.98	0.74
Median	0.42	1.59	0.56
Std. Dev.	0.23	1.25	0.61
Minimum	0.10	0.35	0.36
Maximum	1.71	5.60	2.66

4.1.2 Ratios by group

Table 3: Descriptive statistics for planar ratios for the 4 groups of the chest analysis

Group		Ratio GM 10” lung / 10” myocardium	Ratio GM 10” lung / 10” arm	Ratio GM 60” lung / 60” myocardium	Ratio GM 60”lung / 60” arm	lung retention ratio GM 60 lung / 10 lung
KS in lungs	Mean	0.56	4.00	1.28	2.95	0.98
	Median	0.54	3.30	1.20	2.55	0.85
	Minimum	0.27	1.09	0.52	1.02	0.62
	Maximum	0.82	10.28	3.28	9.70	2.05
	Std. Dev.	0.13	2.39	0.51	1.62	0.34
	N	36	36	36	36	36
KS not in lungs	Mean	0.55	3.56	1.26	2.92	1.04
	Median	0.55	3.29	1.05	1.92	0.89
	Minimum	0.37	1.51	0.82	1.49	0.61
	Maximum	0.69	8.70	2.15	6.04	2.25
	Std. Dev.	0.09	2.11	0.46	1.69	0.47
	N	11	11	11	11	11

Table 3 cont.

KS other lung pathology	Mean	0.618	3.89	1.29	2.80	0.79
	Median	0.59	3.57	1.23	2.81	0.75
	Minimum	0.51	1.30	0.89	1.54	0.67
	Maximum	0.91	7.14	2.11	4.33	0.98
	Std. Dev.	0.12	1.78	0.39	0.91	0.11
	N	10	10	10	10	10

No KS other lung pathology	Mean	0.61	3.72	0.55	3.06	0.92
	Median	0.61	3.66	0.55	2.70	0.89
	Minimum	0.43	1.05	0.39	2.07	0.68
	Maximum	0.75	6.65	0.67	5.20	1.29
	Std. Dev.	0.18	1.20	0.07	1.02	0.19
	N	15	15	15	15	15

Total	Mean	0.58	3.86	1.16	2.95	0.95
	Median	0.58	3.47	1.07	2.59	0.86
	Minimum	0.27	1.05	0.39	1.02	0.61
	Maximum	0.91	10.28	3.28	9.70	2.25
	Std. Dev.	0.12	2.04	0.52	1.42	0.32
	N	72	72	72	72	72

Table 4: Descriptive statistics for SPECT ratios by group

Group		SPECT ratio lung/ myocardium	SPECT ratio lesion/ lung	SPECT ratio lesion/ myocardium
KS in lungs	Mean	0.409	1.82	0.47
	Median	0.39	1.83	0.46
	Minimum	0.10	1.27	0.36
	Maximum	1.00	2.82	0.66
	Std. Dev.	0.16	0.52	0.11
	N	36	7	7
KS not in lungs	Mean	0.53	0.97	0.44
	Median	0.44	0.97	0.44
	Minimum	0.36	0.97	0.44
	Maximum	1.2	0.97	0.44
	Std. Dev.	0.25	0	0
	N	11	1	1
KS other lung pathology	Mean	0.42	1.45	0.58
	Median	0.40	1.45	0.58
	Minimum	0.34	1.45	0.58
	Maximum	0.60	1.45	0.58
	Std. Dev.	0.09	0	0
	N	7	1	1

Table 4

continued

no KS other lung pathology	Mean	0.55	2.36	1.08
	Median	0.45	1.42	0.67
	Minimum	0.25	0.35	0.37
	Maximum	1.71	5.60	2.66
	Std. Dev.	0.38	1.79	0.83
	N	13	7	7

Total	Mean	0.46	1.98	0.74
	Median	0.42	1.59	0.56
	Minimum	0.10	0.35	0.36
	Maximum	1.71	5.60	2.66
	Std. Dev.	0.23	1.25	0.61
	N	67	16	16

4.1.3 Ratios by KS status

Table 5: Descriptive statistics for planar ratios by KS status

KS		Ratio GM 10” lung/10” myocardium	Ratio GM 10”lung/10” muscle	Ratio GM 60”lung/60” myocardium	Ratio GM 60”lung/60” muscle	Lung retention ratio GM 60” lung/10”lung
positive	Mean	0.57	3.90	1.29	2.92	0.96
	Median	0.55	3.40	1.19	2.54	0.84
	Minimum	0.27	1.09	0.52	1.02	0.61
	Maximum	0.91	10.28	3.28	9.70	2.25
	Std. Dev.	0.12	2.21	0.48	1.51	0.35
	N	57	57	57	57	57
negative	Mean	0.61	3.72	0.55	3.06	0.92
	Median	0.61	3.66	0.55	2.70	0.89
	Minimum	0.43	1.05	0.39	2.07	0.68
	Maximum	0.75	6.65	0.67	5.20	1.29
	Std. Dev.	0.18	1.20	0.071	1.02	0.19
	N	15	15	15	15	15
Total	Mean	0.58	3.86	1.14	2.95	0.95
	Median	0.58	3.47	1.07	2.59	0.86
	Minimum	0.27	1.05	0.39	1.02	0.61
	Maximum	0.91	10.28	3.28	9.70	2.25
	Std. Dev.	0.12	2.04	0.52	1.42	0.32
	N	72	72	72	72	72

Table 6: Descriptive statistics for SPECT ratios by KS status

KS		SPECT Ratio Lung/ Myocardium	SPECT Ratio Lesion/ Lung	SPECT Ratio Lesion/ Myocardium
Positive	Mean	0.43	1.68	0.48
	Median	0.41	1.72	0.46
	Minimum	0.10	0.97	0.36
	Maximum	1.21	2.83	0.66
	Std. Dev.	0.179	0.54	0.10
	N	54	9	9
Negative	Mean	0.55	2.36	1.08
	Median	0.45	1.42	0.67
	Minimum	0.25	0.35	0.37
	Maximum	1.71	5.60	2.66
	Std. Dev.	0.39	1.79	0.83
	N	13	7	7
Total	Mean	0.46	1.98	0.74
	Median	0.42	1.59	0.56
	Minimum	0.10	0.35	0.36
	Maximum	1.71	5.60	2.66
	Std. Dev.	0.23	1.25	0.61
	N	67	16	16

4.1.4 Comparison of ratios between specific groups

Groups:

Group 1 KS with KS-lung involvement

Group 2 KS with no lung involvement

Group 3 KS with other lung pathology

Group 4 Non-KS with other lung pathology

4.1.4 (i) HIV-positive KS patients: lung involvement versus no lung involvement (Groups 1 and 3 vs. 2)

Table 7: Mann-Whitney tests for comparison of median ratios between group 1 and 3 vs. group 2

	Groups 1 and 3 vs. 2	N	Mean Rank	Sum of Ranks	p value
Ratio 10” lung / 10” Myocardium	KS in lungs and KS other pathology	46	29.35	1350.00	0.746
	KS not in lungs	11	27.55	303.00	
	Total	57			
Ratio 10” lung / 10” muscle	KS in lungs and KS other pathology	46	29.63	1363.00	0.558
	KS not in lungs	11	26.36	290.00	
	Total	57			
Ratio 60” lung / 60” myocardium	KS in lungs and KS other pathology	46	29.57	1360.00	0.599
	KS not in lungs	11	26.64	293.00	
	Total	57			

Table 7

continued

Ratio 60” lung / 60” muscle	KS in lungs and KS other pathology	46	29.63	1363.00	0.558
	KS not in lungs	11	26.36	290.00	
	Total	57			
Lung retention ratio 60” lung / 10” lung	KS in lungs and KS other pathology	46	28.41	1307.00	0.585
	KS not in lungs	11	31.45	346.00	
	Total	57			
SPECT Ratio Lung/Myocardium	KS in lungs and KS other pathology	43	25.33	1089.00	0.045
	KS not in lungs	11	36.00	396.00	
	Total	54			
SPECT Ratio Lesion/Lung	KS in lungs and KS other pathology	8	5.50	44.00	0.222
	KS not in lungs	1	1.00	1.00	
	Total	9			
SPECT Ratio Lesion/Myocardium	KS in lungs and KS other pathology	8	5.13	41.00	0.889
	KS not in lungs	1	4.00	4.00	
	Total	9			

At the alpha level of 0.00625 there were no significant associations in Table 7.

4.1.4. (ii) HIV-positive KS patients: KS lung involvement versus other pathology (Group 1 vs. Group 3)

Table 8 a: Mann-Whitney tests for comparison of median ratios between group 1 vs. group 3 using planar imaging

	Groups 1 vs. 3	N	Mean Rank	Sum of Ranks	P value
Ratio 10” lung / 10” myocardium	KS in lungs	36	22.36	805.00	0.286
	KS other pathology	10	27.60	276.00	
	Total	46			
Ratio 10” lung / 10” muscle	KS in lungs	36	23.17	834.00	0.763
	KS other pathology	10	24.70	247.00	
	Total	46			
Ratio 60” lung / 60” myocardium	KS in lungs	36	23.19	835.00	0.783
	KS other pathology	10	24.60	246.00	
	Total	46			
Ratio 60” lung / 60” muscle	KS in lungs	36	23.25	837.00	0.824
	KS other pathology	10	24.40	244.00	
	Total	46			
Lung retention ratio 60” lung / 10” lung	KS in lungs	36	25.03	901.00	0.149
	KS other pathology	10	18.00	180.00	
	Total	46			

Table 8 b: Mann-Whitney tests for comparison of median ratios between group 1 vs. group 3 using SPECT imaging

SPECT Ratio Lung/Myocardium	KS in lungs	36	21.44	772.00	0.529
	KS other pathology	7	24.86	174.00	
	Total	43			
SPECT Ratio Lesion/Lung	KS in lungs	7	4.71	33.00	0.750
	KS other pathology	1	3.00	3.00	
	Total	8			
SPECT Ratio Lesion/Myocardium	KS in lungs	7	4.14	29.00	0.500
	KS other pathology	1	7.00	7.00	
	Total	8			

At the alpha level of 0.00625 there were no significant associations in Table 8.

4.1.4. (iii) HIV- positive KS patients: KS in lungs versus normal lungs (Group 1 vs. Group 2)

Table 9 a: Mann-Whitney tests for comparison of median ratios between group 1 vs. group 2 for planar images

	Group 1 vs. 2	N	Mean Rank	Sum of Ranks	P value
Ratio 10” lung / 10” myocardium	KS in lungs	36	24.06	866.00	0.970
	KS not in lungs	11	23.82	262.00	
	Total	47			
Ratio 10” lung / 10” muscle	KS in lungs	36	24.53	883.00	0.647
	KS not in lungs	11	22.27	245.00	
	Total	47			
Ratio 60” lung / 60” myocardium	KS in lungs	36	24.53	883.00	0.647
	KS not in lungs	11	22.27	245.00	
	Total	47			
Ratio 60” lung / 60” muscle	KS in lungs	36	24.56	884.00	0.629
	KS not in lungs	11	22.18	244.00	
	Total	47			
Lung retention ratio 60” lung / 10” lung	KS in lungs	36	23.75	855.00	0.833
	KS not in lungs	11	24.82	273.00	
	Total	47			

Table 9 b: Mann-Whitney tests for comparison of median ratios between group 1 vs. group 2 for SPECT images

SPECT Ratio Lung/Myocardium	KS in lungs	36	21.86	787.00	0.054
	KS not in lungs	11	31.00	341.00	
	Total	47			
SPECT Ratio Lesion/Lung	KS in lungs	7	5.00	35.00	0.250
	KS not in lungs	1	1.00	1.00	
	Total	8			
SPECT Ratio Lesion/Myocardium	KS in lungs	7	4.57	32.00	1.000
	KS not in lungs	1	4.00	4.00	
	Total	8			

At the alpha level of 0.00625 there were no significant associations in Table 9.

4.1.4 (iv) HIV-positive patients: KS in the lung versus other pathology (Group 1 vs. Groups 3 and 4)

Table 10 a: Mann-Whitney tests for comparison of median ratios between group 1 vs. groups 3 and 4 for planar imaging.

	Group 1 vs. 3 and 4	N	Mean Rank	Sum of Ranks	P value
Ratio 10” lung / 10” myocardium	KS in lungs	36	27.94	1006.00	0.107
	Other pathology	25	35.40	885.00	
	Total	61			
Ratio 10” lung / 10” muscle	KS in lungs	36	29.97	1079.00	0.587
	Other pathology	25	32.48	812.00	
	Total	61			
Ratio 60” lung / 60” myocardium	KS in lungs	36	37.81	1361.00	<0.001*
	Other pathology	25	21.20	530.00	
	Total	61			
Ratio 60” lung / 60” muscle	KS in lungs	36	29.56	1064.00	0.446
	Other pathology	25	33.08	827.00	
	Total	61			
Lung retention ratio 60” lung / 10” lung	KS in lungs	36	32.50	1170.00	0.428
	Other pathology	25	28.84	721.00	
	Total	61			

* statistically significant at 0.00625 level

Table 10 b: Mann-Whitney tests for comparison of median ratios between group 1 vs. groups 3 and 4 for SPECT imaging.

SPECT Ratio Lung/Myocardium	KS in lungs	36	26.32	947.50	0.179
	Other pathology	20	32.42	648.50	
	Total	56			
SPECT Ratio Lesion/Lung	KS in lungs	7	8.14	57.00	0.955
	Other pathology	8	7.88	63.00	
	Total	15			
SPECT Ratio Lesion/Myocardium	KS in lungs	7	5.29	37.00	0.029
	Other pathology	8	10.38	83.00	
	Total	15			

The ratio of 60” lung to heart was statistically significantly different between the comparison groups (Group 1 vs. groups 3 and 4) where $p<0.001$.

4.1.4 (v) HIV- positive patients: lung involvement versus normal lungs (Groups 1,3 and 4 vs. Group 2)

Table 11 a: Mann-Whitney tests for comparison of median ratios between groups 1, 3 and 4 vs. group 2 for planar imaging.

	Group 1, 3 and 4 vs. 2	N	Mean Rank	Sum of Ranks	P value
Ratio 10” lung / 10” myocardium	any lung involvement	61	37.36	2279.00	0.411
	normal lungs	11	31.73	349.00	
	Total	72			
Ratio 10” lung / 10” muscle	any lung involvement	61	37.28	2274.00	0.457
	normal lungs	11	32.18	354.00	
	Total	72			
Ratio 60” lung / 60” myocardium	any lung involvement	61	35.57	2170.00	0.377
	normal lungs	11	41.64	458.00	
	Total	72			
Ratio 60” lung / 60” muscle	any lung involvement	61	37.36	2279.00	0.411
	normal lungs	11	31.73	349.00	
	Total	72			
Lung retention ratio 60” lung / 10” lung	any lung involvement	61	36.03	2198.00	0.656
	normal lungs	11	39.09	430.00	
	Total	72			

Table 11 b: Mann-Whitney tests for comparison of median ratios between groups 1, 3 and 4 vs. group 2 for SPECT imaging.

SPECT Ratio Lung/Myocardium	any lung involvement	56	32.34	1811.00	0.115
	normal lungs	11	42.45	467.00	
	Total	67			
SPECT Ratio Lesion/Lung	any lung involvement	15	8.93	134.00	0.250
	normal lungs	1	2.00	2.00	
	Total	16			
SPECT Ratio Lesion/Myocardium	any lung involvement	15	8.73	131.00	0.625
	normal lungs	1	5.00	5.00	
	Total	16			

At the alpha level of 0.00625 there were no significant associations in Table 11

Table 11a and 11b show that there were no significant differences between the ratios in these subgroups.

4.1.4 (vi) HIV- positive patients: KS in lung versus all others (Group 1 vs. Groups 2, 3 and 4)

Table 12 a: Mann-Whitney tests for comparison of median ratios between group 1 vs. groups 2, 3 and 4 for planar imaging.

	Group 1 vs. 2,3, and 4	N	Mean Rank	Sum of Ranks	P value
Ratio 10” lung / 10” myocardium	KS in lungs	36	33.50	1206.00	0.224
	all others	36	39.50	1422.00	
	Total	72			
Ratio 10” lung / 10” muscle	KS in lungs	36	36.00	1296.00	0.839
	all others	36	37.00	1332.00	
	Total	72			
Ratio 60” lung / 60” myocardium	KS in lungs	36	43.83	1578.00	0.003*
	all others	36	29.17	1050.00	
	Total	72			
Ratio 60” lung / 60” muscle	KS in lungs	36	35.61	1282.00	0.179
	all others	36	37.39	1346.00	
	Total	72			
Lung retention ratio 60” lung / 10” lung	KS in lungs	36	37.75	1359.00	0.612
	all others	36	35.25	1269.00	
	Total	72			

Table 12 b: Mann-Whitney tests for comparison of median ratios between group 1 vs. groups 2, 3 and 4 for SPECT imaging.

SPECT Ratio Lung/Myocardium	KS in lungs	36	29.68	1068.50	0.051
	all others	31	39.02	1209.50	
	Total	67			
SPECT Ratio Lesion/Lung	KS in lungs	7	9.14	64.00	0.681
	all others	9	8.00	72.00	
	Total	16			
SPECT Ratio Lesion/Myocardium	KS in lungs	7	5.86	41.00	0.055
	all others	9	10.56	95.00	
	Total	16			

* statistically significant at 0.00625 level

The ratio 60” heart to lung was statistically significantly different in the subgroups compared in Table 12 (p=0.003). The ratio was higher in the group with KS in the lungs.

4.1.4 (vii) HIV- positive patients: KS versus non-KS (Group 1, 2, and 3 vs. 4)

Table 13 a: Mann-Whitney tests for comparison of median ratios between group 1, 2, and 3 vs. group 4 for planar imaging.

	KS	N	Mean Rank	Sum of Ranks	p value
Ratio 10” lung / 10” myocardium	positive	57	34.51	1967.00	0.116
	negative	15	44.07	661.00	
	Total	72			
Ratio 10” lung / 10” muscle	positive	57	35.71	2035.50	0.533
	negative	15	39.50	592.50	
	Total	72			
Ratio 60” lung / 60” myocardium	positive	57	43.75	2494.00	<0.001*
	negative	15	8.93	134.00	
	Total	72			
Ratio 60” lung / 60” muscle	positive	57	35.04	1997.50	0.250
	negative	15	42.03	630.50	
	Total	72			
Lung retention ratio 60” lung / 10” lung	positive	57	35.96	2049.50	0.667
	negative	15	38.57	578.50	
	Total	72			

Table 13 b: Mann-Whitney tests for comparison of median ratios between group 1, 2, and 3 vs. group 4 for SPECT imaging.

SPECT Ratio Lung/ Myocardium	positive	54	32.71	1766.50	0.270
	negative	13	39.35	511.50	
	Total	67			
SPECT Ratio Lesion/Lung	positive	9	8.22	74.00	0.837
	negative	7	8.86	62.00	
	Total	16			
SPECT Ratio Lesion/ Myocardium	positive	9	6.22	56.00	0.031
	negative	7	11.43	80.00	
	Total	16			

* statistically significant at the 0.00625 level.

The ratio 60” heart to lung was statistically significantly different in the KS negative and positive groups ($p<0.001$). The ratio was higher in the group with KS.

4.1.4. (viii) HIV-positive patients: KS in the lungs vs. non KS other lung pathology (Group 1 vs. Group 4)

Table 14 a: Mann-Whitney tests for comparison of median ratios between group 1 vs. group 4 for planar imaging.

	Group	N	Mean Rank	Sum of Ranks	P value
Ratio 10” lung / 10” myocardium	KS in lungs	36	24.08	867.00	0.154
	no KS other lung pathology	15	30.60	459.00	
	Total	51			
Ratio 10” lung / 10” muscle	KS in lungs	36	25.31	911.00	0.605
	no KS other lung pathology	15	27.67	415.00	
	Total	51			
Ratio 60” lung / 60” myocardium	KS in lungs	36	33.11	1192.00	<0.001*
	no KS other lung pathology	15	8.93	134.00	
	Total	51			
Ratio 60” lung / 60” muscle	KS in lungs	36	24.81	893.00	0.374
	no KS other lung pathology	15	28.87	433.00	
	Total	51			
Lung retention ratio 60” lung / 10” lung	KS in lungs	36	25.97	935.00	0.984
	no KS other lung pathology	15	26.07	391.00	
	Total	51			

Table 14 b: Mann-Whitney tests for comparison of median ratios between group 1 vs. group 4 for SPECT imaging.

SPECT Ratio Lung/Myocardium	KS in lungs	36	23.38	841.50	0.185
	no KS other lung pathology	13	29.50	383.50	
	Total	49			
SPECT Ratio Lesion/Lung	KS in lungs	7	7.43	52.00	1.000
	no KS other lung pathology	7	7.57	53.00	
	Total	14			
SPECT Ratio Lesion/ Myocardium	KS in lungs	7	5.14	36.00	0.038
	no KS other lung pathology	7	9.86	69.00	
	Total	14			

* statistically significant at the 0.00625 level.

The ratio 60” heart to lung was statistically significantly different between the two groups compared in the above table ($p<0.001$). The ratio was higher in the group with KS in the lungs.

4.1.5. Demographics

There was no significant difference between the distribution of males and females in the four groups (p=0.766). Table 15 shows the cross-tabulation with row percentages.

Table 15: Cross-tabulation of gender by group

		SEX		Total
Group		F	M	
KS in lungs	Count	17	19	36
	% within Group	47.2%	52.8%	100.0%
KS not in lungs	Count	5	6	11
	% within Group	45.5%	54.5%	100.0%
KS other lung pathology	Count	6	4	10
	% within Group	60.0%	40.0%	100.0%
No KS other lung pathology	Count	9	6	15
	% within Group	60.0%	40.0%	100.0%
Total	Count	37	35	72
	% within Group	51.4%	48.6%	100.0%

P=0.766

Ages ranged from 20 to 75 years with a mean age of 36.8 and a standard deviation of 10.4 years. Table 16 shows the descriptive statistics for age by group. The mean age of the non KS group was higher than the other groups.

Table 17 shows that this difference in mean ages between the groups was statistically significant (p=0.008). The post hoc tests (Table 18) show that differences were found between the non KS group and the group with KS in the lung and KS other lung pathology.

Table 16: Descriptive statistics for age by group (all ages given in years)

Group	Mean	Minimum	Maximum	Std. Dev.	N
KS in lungs	34.86	20	55	8.28	36
KS not in lungs	37.82	27	52	7.55	11
KS other lung pathology	31.50	20	52	9.35	10
no KS other lung pathology	44.07	28	75	13.86	15
Total	36.76	20	75	10.40	72

Table 17: ANOVA table for comparison of mean age by group

	Sum of Squares	df	Mean Square	F	p value
Between Groups	1219.61	3	406.54	4.29	0.008
Within Groups	6445.38	68	94.79		
Total	7664.99	71			

Table 18: Post hoc Bonferroni multiple comparison tests for comparison of mean age between the four groups (all age values given in years)

(I) Group	(J) Group	Difference (I-J)	Std. Error	Sig.	95% Confidence Interval for diff.	
KS in lungs	KS not in lungs	-2.96	3.35	1.000	-12.07	6.16
	KS other lung pathology	3.36	3.48	1.000	-6.10	12.82
	no KS other lung pathology	-9.21(*)	2.99	0.018	-17.34	-1.07
KS not in lungs	KS in lungs	2.96	3.35	1.000	-6.16	12.07
	KS other lung pathology	6.32	4.25	0.853	-5.24	17.88
	no KS other lung pathology	-6.25	3.87	0.663	-16.75	4.25
KS other lung pathology	KS in lungs	-3.36	3.48	1.000	-12.82	6.10
	KS not in lungs	-6.32	4.25	0.853	-17.88	5.24
	no KS other lung pathology	-12.57(*)	3.98	0.014	-23.37	-1.77
no KS other lung pathology	KS in lungs	9.21(*)	2.99	0.018	1.07	17.34
	KS not in lungs	6.25	3.87	0.663	-4.25	16.75
	KS other lung pathology	12.57(*)	3.98	0.014	1.77	23.37

* The mean difference is significant at the 0.05 level.

4.1.6. Follow-up post-treatment

Four subjects were followed up to post treatment. Changes in mean and median values between pre- and post-treatment are shown in Tables 19 to 22. Due to the small sample size purely descriptive analysis was done on the change between pre and post treatment in these four subjects.

Table 19: Pre- and post-treatment (Rx) 10” lung, arm and myocardium counts

		Pre Rx 10” muscle	Post Rx 10” muscle	Pre Rx 10” myocardium	Post Rx 10” myocardium	Pre Rx 10” lung	Post Rx 10” lung
N	Valid	4	4	4	4	4	4
Mean		5.85	4.39	32.03	35.91	17.25	17.49
Median		4.80	4.72	30.77	32.85	19.17	17.46
Std. Dev.		3.32	2.25	17.81	8.03	8.90	8.54
Minimum		3.16	1.66	11.86	30.16	4.90	7.22
Maximum		10.63	6.45	54.74	47.79	25.75	27.83

Table 20: Pre- and post-treatment (Rx) 60” lung, arm and myocardium counts

		Pre Rx 60” muscle	Post Rx 60” muscle	Pre Rx 60” lung	Post Rx 60” lung	Pre Rx 60” myocardium	Post Rx 60” myocardium
N	Valid	4	4	4	4	4	4
Mean		6.56	5.96	16.25	15.05	14.99	36.41
Median		6.54	6.60	15.10	14.25	12.66	33.72
Std. Dev.		3.40	1.92	5.43	4.81	8.81	7.64
Minimum		2.45	3.26	11.00	10.35	7.06	30.66
Maximum		10.69	7.39	23.79	21.38	27.59	47.56

Table 21: Pre- and post-treatment (Rx)10” and 60” ratios with lung versus myocardium ratios given as lung /myocardium and lung muscle ratios given as lung /muscle.

	Pre Rx 10”lung /myocrd	Post Rx 10”lung /myocrd	Pre Rx 60”lung /myocrd	Post Rx 60”lung /myocrd	Pre Rx 10”lung /muscle	Post Rx 10”lung /muscle	Pre Rx 60”lung /muscle	Post Rx 60”lung /muscle
N Valid	4	4	4	4	4	4	4	4
Mean	0.53	0.48	1.20	0.41	3.05	4.13	2.85	2.62
Median	0.54	0.55	1.19	0.43	2.82	4.45	2.46	2.79
Std. Dev.	0.11	0.18	0.29	0.08	1.47	0.77	1.13	0.63
Minimum	0.41	0.22	0.86	0.30	1.55	2.99	1.99	1.72
Maximum	0.64	0.60	1.56	0.49	5.01	4.63	4.49	3.17

Table 22: Pre- and post- treatment (Rx) lung retention ratios

	Pre Rx GM 60” lung/ 10” lung Retention Ratio	Post Rx GM 60” lung/ 10” lung Retention Ratio
N Valid	4	4
Mean	1.19	0.97
Median	0.92	0.89
Std. Dev.	0.71	0.35
Minimum	0.68	0.65
Maximum	2.25	1.43

4.1.7. CD4 count analysis

CD4 counts were available for 43 participants with KS. The descriptive statistics for each group are shown in Table 23. The majority of participants with CD4 counts were from the group with KS in the lungs (n=28) and their median CD4 count was 185. There were 8 participants with CD4 counts in the KS other lung pathology group and their CD4 counts tended to be the highest of the groups with a median of 217. There was no significant difference in median CD4 count in the three groups (p=0.629).

Table 23: Descriptive statistics for CD4 count by group

Group	Median	Minimum	Maximum	N
KS in lungs	185.00	3	1002	28
KS not in lungs	194.00	117	459	7
KS other lung pathology	217.00	77	1228	8
Total	194.00	3	1228	43

Table 24 shows that CD4 count was not correlated with any of the ratio measurements in all groups.

Table 24: Spearman's correlation between CD4 count and ratios in all groups

		CD4
Ratio GM 10" lung/ 10" myocardium	Corr. Coef.	0.121
	Sig. (2-tailed)	0.439
	N	43
Ratio GM 10" lung / 10" muscle	Corr. Coef.	0.124
	Sig. (2-tailed)	0.427
	N	43
Ratio GM 60" lung / 60" myocardium	Corr. Coef.	0.029
	Sig. (2-tailed)	0.855
	N	43

Table 24 continued		
Ratio GM 60” lung / 60” muscle	Corr. Coef.	0.002
	Sig. (2-tailed)	0.989
	N	43
Lung Retention Ratio GM 60” lung / 10” lung	Corr. Coef.	-0.242
	Sig. (2-tailed)	0.118
	N	43
SPECT Ratio Lung / Myocardium	Corr. Coef.	-0.157
	Sig. (2-tailed)	0.333
	N	40
SPECT Ratio Lesion/ Lung	Corr. Coef.	-0.450
	Sig. (2-tailed)	0.224
	N	9
SPECT Ratio Lesion/ Myocardium	Corr. Coef.	-0.017
	Sig. (2-tailed)	0.966
	N	9

Similarly within group 1 (KS in the lungs) there was no correlation between CD4 counts and ratios (Table 25).

Table 25: Spearman's correlation between CD4 count and ratios in group 1

Ratio GM 10" lung/ 10" myocardium	Corr. Coef.	0.086
	Sig. (2-tailed)	0.662
	N	28
Ratio GM 10" lung / 10" muscle	Corr. Coef.	0.169
	Sig. (2-tailed)	0.390
	N	28
Ratio GM 60" lung / 60" myocardium	Corr. Coef.	0.009
	Sig. (2-tailed)	0.963
	N	28
Ratio GM 60" lung / 60" muscle	Corr. Coef.	-0.030
	Sig. (2-tailed)	0.881
	N	28
Lung Retention Ratio GM 60" lung / 10" lung	Corr. Coef.	-0.273
	Sig. (2-tailed)	0.161
	N	28
SPECT Ratio Lung/ Myocardium	Corr. Coef.	-0.121
	Sig. (2-tailed)	0.540
	N	28
SPECT Ratio Lesion/ Lung	Corr. Coef.	-0.536
	Sig. (2-tailed)	0.215
	N	7
SPECT Ratio Lesion/ Myocardium	Corr. Coef.	-0.071
	Sig. (2-tailed)	0.879
	N	7

4.2 Cutaneous Analysis Results

4.2.1. Lymphoedema

Table 26 shows the results of the comparison between the clinical rating of lymphedema versus MIBI detection. There were 7 false positives and 6 false negatives. The sensitivity was 63% and specificity was higher at 72%.

Table 26: Cross tabulation of lymphoedema clinical vs. MIBI (n=41)

		Lymphoedema clinical		Total
		Yes	No	
Lymphoedema MIBI	Yes	10	23	33
	No	5	3	8
Total		15	26	41

4.2.2. Lymph Nodes

Table 27 shows the clinical and imaging data on detection of lymph nodes. The ^{99m}Tc-MIBI imaging detected more patients with lymph nodes than on clinical examination. The ^{99m}Tc-MIBI images reported only one false negative.

Table 27: Cross tabulation of lymph nodes clinical vs. MIBI (n=43)

		Lymph nodes clinical		Total
		yes	no	
Lymph nodes MIBI	Yes	9	24	33
	No	1	9	10
Total		10	33	43

4.2.3. Extent of Lesions

Table 28: Cross tabulation of extent of lesions clinical vs. MIBI imaging (n=37)

		Extent of lesions: clinical				Total
		Diffuse	Scattered	Isolated	Local	
Extent of lesions:MIBI	No lesions	1	8	0	1	10
	Diffuse	9	0	1	1	11
	Scattered	1	5	1	1	8
	Isolated	0	1	3	1	5
	Local	0	0	0	3	3
Total		11	14	5	7	37

4.2.4 Skin Description

Table 29: Cross tabulation of skin description clinical vs. MIBI (n=43)

	Clinical skin description				Total
MIBI skin description ↓	No lesions	Plaque	Plaque & nodule	Nodule	
No lesions	2	10	1	1	14
Plaque	2	9	2	5	18
Plaque & nodule	0	4	0	1	5
Nodule	0	2	2	2	6
Total	4	25	5	9	43

A detailed discussion of the results follows in Chapter 5.

CHAPTER 5

DISCUSSION

In patients with AIDS, there is often co-existence of pulmonary KS and opportunistic infection with non-specific constitutional symptoms. Prompt detection, diagnosis and treatment can reduce patient morbidity and mortality. The aim of this study was to evaluate ^{99m}Tc-MIBI imaging of KS in AIDS patients.

5.1 Discussion of Chest Data

One of the objectives was to determine the efficacy, in terms of, sensitivity and specificity, of ^{99m}Tc-MIBI imaging in differentiating pulmonary AIDS-KS from opportunistic infection. In AIDS patients there is often coexistence of opportunistic infection and KS (Figure 2). Pulmonary KS is the most serious form of KS with high fatality rate and a median survival time of 3-10 months (Holkova *et al,* 2001:3848). Pulmonary KS can be a serious complication of AIDS-KS patients and often mimics *Pneumocystis carinii* infection.

In HIV and HHV8-seropositive patients who are at risk of developing AIDS-KS, chest radiograph is used to screen for pulmonary involvement (Cannon et al, 2003: 85, Dezube, 2004: 445). Bronchoscopy is reserved for patients with persistent respiratory symptoms or abnormal chest radiograph (Lilenbaum & Ratner, 1994: 142). However, pulmonary or mediastinal involvement is usually a late manifestation of KS and chest radiographs may not be contributory. Because bronchoscopy may also be unreliable for diagnosing endobronchial KS in 30% of patients, open lung biopsy may be required (Lee *et al*, 1991: 409). Because of the decreased incidence of PCP with prophylaxis, there is a shift to opportunistic

infection with TB, mycobacterium avium-intracellulare, cytomegalovirus and cryptococcus. Chest radiograph, lung volume and spirometry measurement are not sensitive or specific (Lee et al, 1994:389; Abdel-Dayem et al 1996:1666). Diagnostic sensitivities of sputum analysis, bronchoscopy with lavage and biopsy, and open-lung biopsy are 56, 98, and 88% respectively (Sittler et al, 1990: 73).

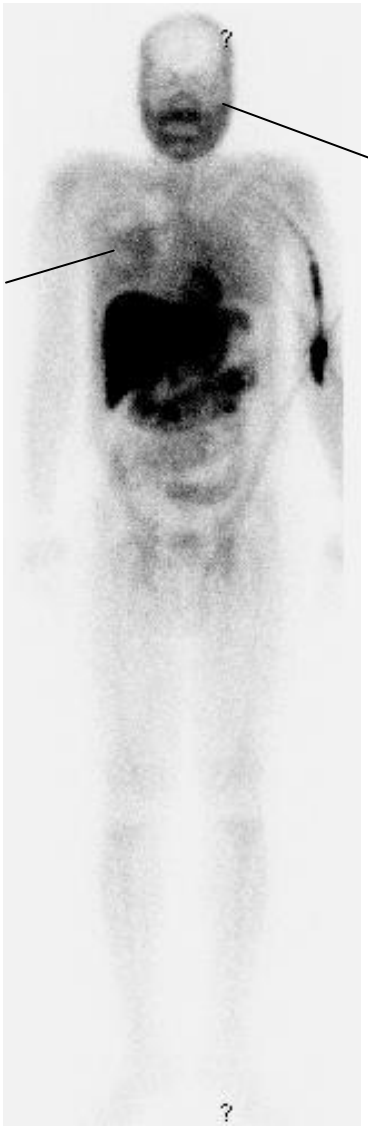


Figure 2: Anterior whole-body ^{99m}Tc-MIBI image demonstrating a KS pulmonary lesion (a) and facial involvement (b) in a 44 year old male patient

In this study, the final patient diagnosis was made by histological, microbiological or bronchoscopic findings. Using the final diagnosis as the gold standard, sensitivity, specificity, and accuracy of the visual interpretation and quantitative analysis methods of the planar (10 minute and 60 minute) and SPECT images of the chest was determined.

All the patients studied were HIV positive. The data obtained from the chest images was divided into 4 groups as shown in figure 1.

Group 1: KS patients with KS lung involvement (n=36)

Group 2: KS patients with no KS lung involvement (n=11)

Group 3: KS patients with other lung pathology (n=10)

Group 4: Non-KS patients with other lung pathology (control group, n=15)

The comparison of the quantitative ratios from the KS patients that had confirmed KS involvement of the lungs (group 1) on bronchoscopy with KS patients and non-KS patients who had other lung pathology (groups 3 and 4) demonstrated statistically significant ($p<0.001$) changes for the 60" lung/myocardium ratios of the geometric mean of the anterior and posterior planar and SPECT images (Table 10). This is also evident from Table 12, where again for 60" lung/myocardium ratios the p value is significant at $p=0.003$ for the comparison between KS patients with lung involvement (group 1) and those with no KS lung involvement (groups 2,3 and 4). The analysis of the 60" lung/myocardium ratios of KS patients (groups 1,2 and 3) with or without lung involvement on Table 13 once more shows statistical significance ($p<0.001$) when compared to non-KS

patients (group 4). Table 14 shows the analysis of group 1, KS in lungs versus group 4, non-KS patients that display other lung pathology. The 60" lung/myocardium ratios of the geometric mean of the anterior and posterior planar images for this group is statistically significant with a p value <0.001.

It is evident from these results that the 60" lung/myocardium ratios of the geometric mean of the anterior and posterior planar images are significant when compared to the 10" lung/myocardium ratios (Table 3). The possible reason for this is that at 10-minutes post-injection of the ^{99m}Tc-MIBI, equilibrium had not been reached. The lung counts are as expected, significantly lower than the heart counts.

When examining the 10-minute and 60-minute lung/muscle ratios, the mean value at 10-minutes (2.94) is much higher than that at 60-minutes (0.58). This is also thought to be due to the fact that equilibrium was not reached at 10 minutes.

What is of note on Table 3 is that the mean value of the 60-minute lung/myocardium ratio for group 4, that is, the non KS group, is much lower (0.55) than the groups with KS patients (1.30,1.26 and 1.30 for groups 1, 2 and 3 respectively). This is further supported in the comparison between KS positive and KS-negative patients where the mean value for the same ratio is 0.55 for the non-KS patients and 1.29 for the KS-positive patients (Table 5).

A comparison of planar ratios for the 60" lung/myocardium and SPECT lung/myocardium shows a total mean value for all groups of 0.46 for SPECT (Table 6) and 1.14 for planar images (Table 5). The SPECT ratio for each group is considerably lower; comparison of 60" lung/myocardium planar to SPECT

ratios for the groups 1,2, 3 and 4 show means values of 1.30,1.26 and 1.30 and 0.55 for planar and 0.41,0.53, 0.42 and 0.55 for SPECT (Tables 3 and 4). SPECT imaging is more sensitive than planar imaging as there are no overlying structures to attenuate the radioactive uptake. From this study it is evident that SPECT imaging is more effective than planar imaging (Figure 3).

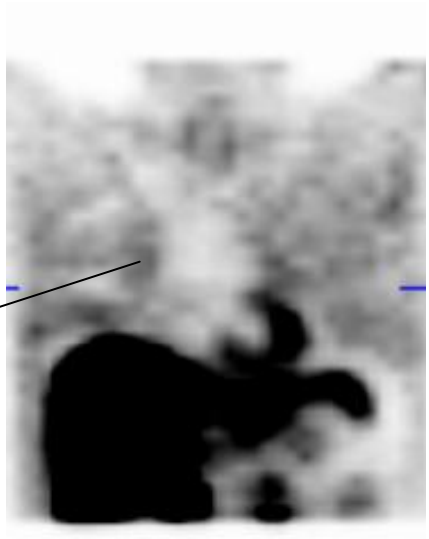


Figure 3: ^{99m}Tc-MIBI SPECT image of the chest showing KS lung lesion (a) in a 44 year- old male patient

The summary of the analysis of the 60" lung/myocardium ratios of the geometric mean of the anterior and posterior planar images is statistically significant in various comparisons as shown in Table 30 (i).

Table 30 (i): Summary of values of the 60” lung/myocardium ratios of the geometric mean of the anterior and posterior planar images that are statistically significant

Table No	Groups	P value	Description
10	1 vs. 3 & 4	<0.001	KS lung involvement vs. KS other lung pathology and non-KS other lung pathology
12	1 vs. 2, 3 & 4	0.003	KS lung involvement vs. no KS lung involvement
13	1, 2 & 3 vs. 4	<0.001	KS patients vs. non-KS patients
14	1 vs. 4	<0.001	KS lung involvement vs. non-KS other lung pathology

The results obtained thus far show that the 60” lung/myocardium ratios of the geometric mean of the anterior and posterior planar ^{99m}Tc-MIBI images may be useful to establish whether there is KS lung involvement.

The summary of the analysis of the various combinations of groups as listed in Table 30(ii) below showed no statistical significance:

Table 30 (ii): Summary of Values that are not statistically significant

Table	Groups	Description
7	1, 3 vs. 2	KS lung involvement and KS other lung pathology vs. no KS lung involvement.
8	1 vs. 3	KS lung involvement vs. KS other lung pathology
9	1 vs. 2	KS lung involvement vs. no KS lung involvement
11	1, 3, 4 vs. 2	KS lung involvement, KS other lung pathology and non-KS other lung pathology vs. no KS lung involvement.

To express the relationship between sensitivity and specificity, receiver operating characteristic (ROC) curves were generated so as to indicate how severe the trade-off is between sensitivity and specificity at any given threshold value.

By definition, the sensitivity (Sn) of a test is the probability that the test is positive when given to a group of patients with the disease. The formula for sensitivity is:

Sn = TP / (TP + FN)

where TP and FN are the number of true positive and false negative results, respectively (Simon, 2004).

The specificity (Sp) of a test is the probability that the test will be negative among patients who do not have the disease. The formula for specificity is:

Sp = TN / (TN + FP)

where TN and FP and the number of true negative and false positive results, respectively (Simon, 2004)

The cut-off points were selected so as to define a true-positive from a true-negative such that the sensitivity and specificity were acceptable. If too high a cut point was selected, the test would be more specific, that is, few false positives, but the sensitivity would be decreased, that is, increase in false negatives. If too low a cut point were selected, it would be more sensitive (fewer false negatives) but less specific, that is, more false positives (Peer, 2001:69).

5.1.1 KS with lung involvement versus other lung pathology (Group 1 vs. Groups 3 and 4)

The 60" lung/myocardium ratio was significantly different between group 1 vs. groups 3 and 4 (Table 10), thus further ROC analysis was done on this ratio. The objective was to find cut-points of the ratios that would optimize sensitivity and specificity for predicting KS in the lung compared with other lung pathology. The ROC analysis curve (Fig 4) of the sensitivity by 1-specificity at each data value (Table A - Appendix 5) reflected an area under the curve of 0.772 that is significantly greater than 0.5 ($p<0.001$) as shown in Table 31. This ratio was thus a good predictor of KS in the lungs compared to the groups with other lung pathology. At the 95% Confidence Interval a sensitivity of 94% and specificity of 60% was obtained for 60" lung/myocardium ratios for KS with lung involvement versus other lung pathology (Table 32).

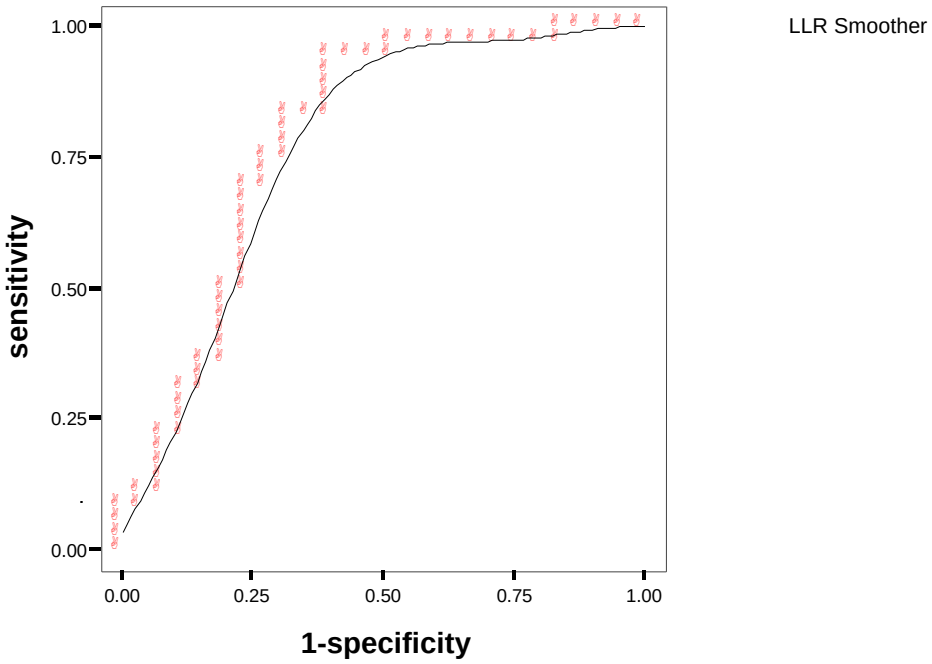


Figure 4: A smooth fitted ROC curve of sensitivity by 1-specificity for 60”lung/myocardium ratios for group 1 vs. Groups 3 and 4 generated using LLR smoother and plotted with the raw data points.

Table 31: Area Under the Curve for 60” lung/myocardium ratio group 1 vs. groups 3 and 4

Area	Std. Error(a)	Asymptotic Sig.(b)	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
0.772	0.067	<0.001	0.641	0.903

a Under the nonparametric assumption

b Null hypothesis: true area = 0.5

The cut point of 0.730 was chosen so as to optimize sensitivity and specificity. For group 1 vs. group 3 and 4, at the selected cut-point of 0.730 for 60“ lung/myocardium ratios, the following screening statistics were obtained (with 95% confidence intervals).

Table 32: Group 1 vs. groups 3 and 4 screening statistics for 60“ lung/ myocardium ratios at cut point of 0.730

Screening Statistics	Statistical Value	95% Confidence Interval Limits
Prevalence	0.59	[0.46, 0.71]
Sensitivity	0.94	[0.80, 0.99]
Specificity	0.60	[0.39, 0.78]
Accuracy	0.80	[0.68, 0.89]
Predictive value of +ve result	0.77	[0.62, 0.88]
Predictive value of -ve result	0.88	[0.62, 0.98]

The Positive Predictive Value (PPV) of a test is the probability that the patient has the disease when restricted to those patients who test positive.

$$PPV = TP / (TP + FP)$$

where TP and FP are the number of true positive and false positive results, respectively (http://www.cmh.edu/stats/definitions)**.**

The Negative Predictive Value (NPV) of a test is the probability that the patient will not have the disease when restricted to all patients who test negative.

$$NPV = TN / (TN + FN)$$

where TN and FN are the number of true negative and false negative results, respectively (http://www.cmh.edu/stats/definitions)**.**

5.1.2 KS with lung involvement versus KS with no lung involvement, KS with other lung pathology and non-KS with other lung pathology (Group 1 vs. Groups 2,3 and 4)

In the analysis comparing the group with KS lung involvement to all the other groups (group 1 vs. groups 2, 3 and 4) the 60" lung/myocardium ratio was significant at p = 0.003 (Table 12).

Figure 5 shows the ROC curve of the sensitivity by 1-specificity at each data value. The area under the curve (0.704) was significantly greater than 0.5 (p=0.003) thus this ratio was a good predictor of KS in the lungs relative to the other pathology groups (Table 33).

Table B in Appendix 5, shows the sensitivity and 1-specificity of each of the data points. The sensitivity was 86% and specificity 50% at the 95% confidence

interval (Table 34) using the 60”lung/myocardium ratios in the comparison of the groups of KS lung involvement to all the other groups (group 1 vs. groups 2, 3 and 4).

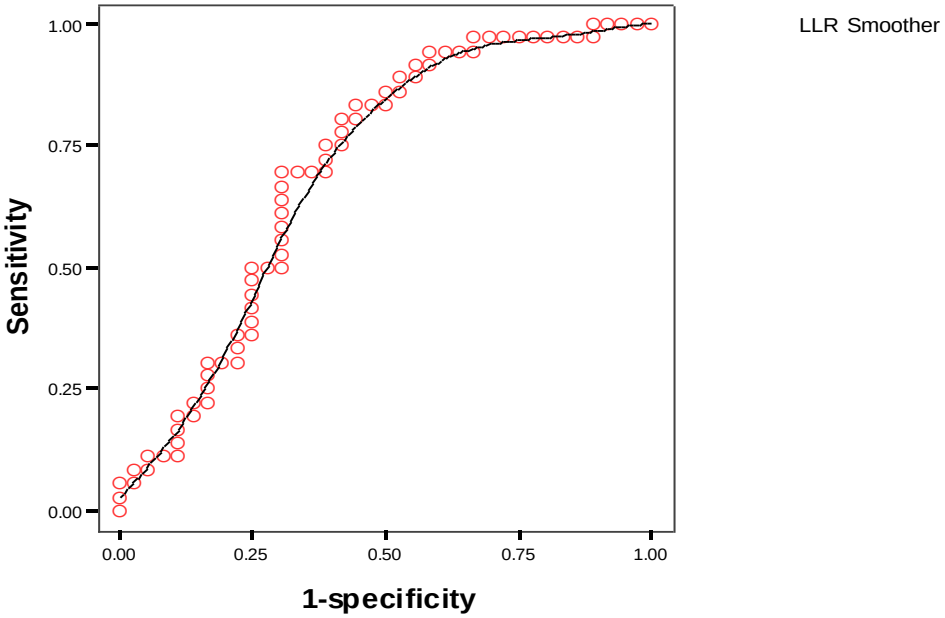


Figure 5: A smooth fitted ROC curve of sensitivity by 1-specificity for 60” lung/myocardium ratio for Group 1 vs. Groups 2, 3 and 4 generated using LLR smoother and plotted with the raw data points.

Table 33: Area Under the Curve for 60” lung/myocardium ratio for group1 vs. groups 2, 3 and 4

Area	Std. Error(a)	Asymptotic Sig.(b)	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
0.704	0.063	0.003	0.580	0.827

a Under the nonparametric assumption

b Null hypothesis: true area = 0.5

The cut point of 0.8722 was chosen for group 1 vs. groups 2, 3 and 4, with the following screening statistics:

Table 34: Group 1 vs. groups 2, 3 and 4 screening Statistics for 60” lung/ myocardium ratio at cut point of 0.872

Screening Statistics	Statistical Value	95% Confidence Interval Limits
Prevalence	0.50	[0.38, 0.62]
Sensitivity	0.86	[0.70, 0.95]
Specificity	0.50	[0.33, 0.67]
Accuracy	0.68	[0.56, 0.78]
Predictive value of +ve result	0.63	[0.48, 0.76]
Predictive value of -ve result	0.78	[0.56, 0.92]

5.1.3 KS versus Non- KS other pathology (Group 1, 2 and 3 vs. Group 4)

The 60” lung/myocardium ratio was significantly different ($p<0.001$) between groups 1, 2 and 3 vs. group 4 (Table 13), thus further ROC analysis was done on this ratio. The objective was to find cut-points of the ratios that would optimize sensitivity and specificity for predicting KS vs. non-KS with other lung pathology. The ROC analysis curve (Figure 6) of the sensitivity by 1-specificity at each data value (Table C - Appendix 5) reflected an area under the curve of 0.984 that is significantly greater than 0.5 ($p<0.001$) as shown in Table 35. This ratio was thus a good predictor of KS in the lungs compared to the groups with other lung pathology. At the 95% Confidence Interval, a sensitivity of 98% and specificity of 80% were obtained using 60“ lung/myocardium ratios for KS patients with/without lung involvement versus non-KS with other lung pathology (Table 36).

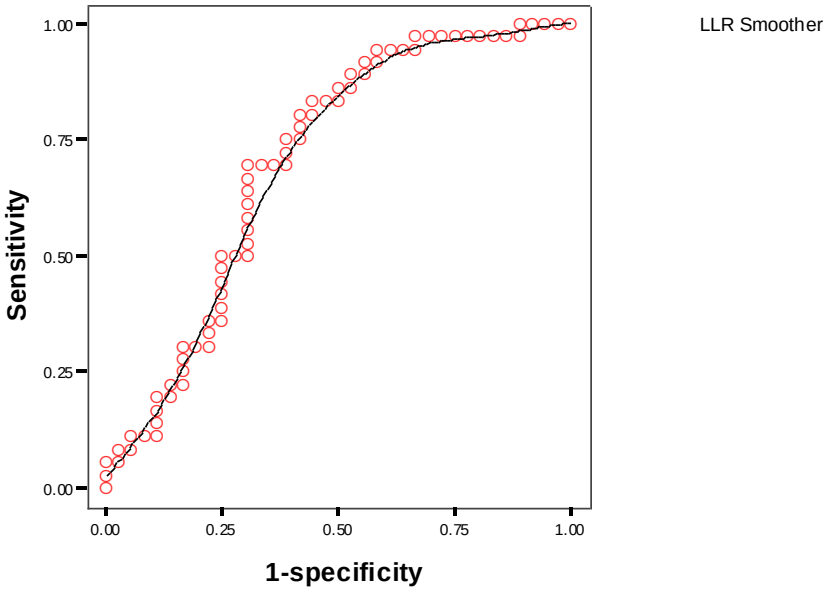


Figure 6: A smooth fitted ROC curve of sensitivity by 1-specificity for 60"lung/myocardium ratio for Groups 1, 2 and 3 vs. 4 generated using LLR smoother and plotted with the raw data points.

Table 35 shows that the 60" lung/myocardium ratio was a significant predictor of differentiating KS from non-KS.

Table 35: Area Under the Curve for 60” lung/myocardium ratio for groups 1, 2 and 3 vs. 4

Area	Std. Error(a)	Asymptotic Sig.(b)	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
0.984	0.014	<0.001	0.957	1.011

a Under the nonparametric assumption

b Null hypothesis: true area = 0.5

At a cut point of 0.614, groups 1, 2 and 3 vs. 4 the following statistics were found:

Table 36: Groups 1, 2 and 3 vs. group 4 screening statistics for 60” lung/myocardium ratios at a cut point of 0.614

Screening Statistics	Statistical Value	95% Confidence Interval Limits
Prevalence	0.79	[0.68, 0.88]
Sensitivity	0.98	[0.89, 1.00]
Specificity	0.80	[0.51, 0.95]
Accuracy	0.94	[0.86, 0.98]
Predictive value of +ve result	0.95	[0.85, 0.99]
Predictive value of -ve result	0.92	[0.62, 1.00]

5.1.4 KS with lung involvement versus Non-Ks other lung pathology (Group 1 vs. Group 4)

In the analysis comparing the group with KS lung involvement with the non-KS group (group 1 vs. group 4) the 60” lung/myocardium ratio was significant at $p<0.001$ (Table 14). Figure 7 shows the ROC curve of the sensitivity by 1-specificity at each data value. The area under the curve (0.974) was significantly greater than 0.5 ($p<0.001$) thus this ratio was a good predictor of KS in the lungs relative to the non-KS group with other lung pathology (Table 37).

Table D in Appendix 5, shows the sensitivity and 1-specificity of each of the data points. The sensitivity was 97% and specificity 80% at the 95% Confidence Interval (Table 38) using the 60”lung/myocardium ratios in the comparison of the groups of KS lung involvement to the non-KS with other lung pathology group (group 1 vs. group 4)

The 60” lung/myocardium ratio was a significantly good predictor for distinguishing KS in the lungs from other non-KS pathology in the lungs as shown in Table 37.

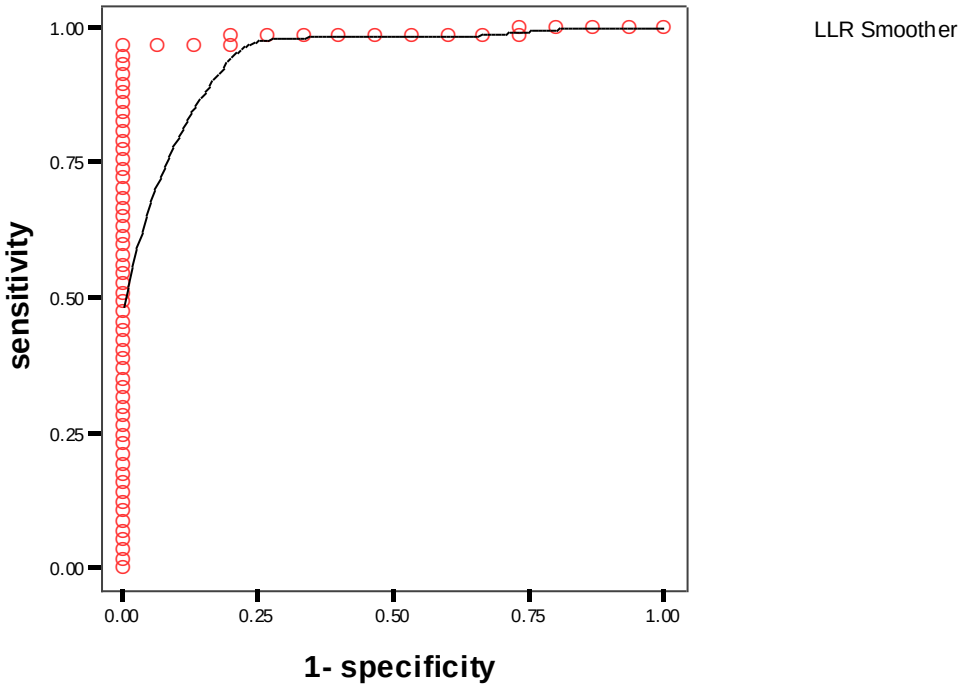


Figure 7: A smooth fitted ROC curve of sensitivity by 1-specificity for 60"lung/myocardium ratio for Group 1 vs. Group 4 generated using LLR smoother and plotted with the raw data points.

Table 37: Area Under the Curve for 60"lung/myocardium ratio for group 1 vs. group 4

Area	Std. Error(a)	Asymptotic Sig.(b)	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
0.974	0.022	<0.001	0.932	1.016

a Under the nonparametric assumption

b Null hypothesis: true area = 0.5

Table 38: Group 1 vs. group 4 screening statistics for 60”lung/myocardium ratios at a cut point of 0.614

Screening Statistics	Statistical Value	95% Confidence Interval Limits
Prevalence	0.71	[0.56, 0.82]
Sensitivity	0.97	[0.84, 1.00]
Specificity	0.80	[0.51, 0.95]
Accuracy	0.92	[0.80, 0.97]
Predictive value of +ve result	0.92	[0.78, 0.98]
Predictive value of -ve result	0.92	[0.62, 1.00]

Thus separating group 4 (non-KS with other lung pathology) from the other groups gave higher values of sensitivity and specificity than trying to distinguish group 1 (KS with lung involvement) from the other groups. Excluding non-KS patients with other lung pathology was more sensitive (97%) and specific (80%) than distinguishing KS with lung involvement.

Goswami *et al* in 2000, indicate that anti-TB treatment decreases the sensitivity of the Ga-67 scans. Should this be true for ^{99m}Tc-MIBI uptake, then perhaps this could have contributed to the discrepancy in uptake of ^{99m}Tc-MIBI in KS patients with lung involvement and with no lung involvement.

From the discussions it is clear that ^{99m}Tc-MIBI imaging is not ideal for differentiating pulmonary KS from opportunistic infection in AIDS-KS patients but

could prove useful to differentiate pulmonary infection from malignancy in non-KS-AIDS patients.

In the analysis of the pre- and post- treatment ratios (Table 21), the 60" lung/myocardium ratio of 0.41 for post-treatment is considerably lower than the 60" lung/myocardium pre-treatment ratio of 1.20. A ^{99m}Tc-MIBI scan covers the entire tumour lesion and may show the pharmacokinetics within tumour tissue more accurately than direct sampling of heterogeneous tumour tissue that requires surgical intervention or biopsy. Also the retention of ^{99m}Tc-MIBI in cells has been reported to be dependent on the activity P-gp coded by multidrug resistance gene, which functions as ATP-dependent efflux pump for cytotoxic substances (Kao *et al,* 1997: 249). ^{99m}Tc-MIBI is reported to be a ligand for MDR and the positive tumour uptake of ^{99m}Tc-MIBI is considered a low expression of P-gp. This suggests that ^{99m}Tc-MIBI imaging may be useful to predict or to check response to treatment for KS.

Although ^{99m}Tc-MIBI imaging does not have high sensitivity or specificity for detecting pulmonary KS, it may be useful as a predictive test or follow-up of response of KS to treatment

5.2 Discussion of Cutaneous Data

It was hypothesized (see 1.3.1 and 1.3.2) that ^{99m}Tc-MIBI imaging would be both sensitive and specific for the detection of extracutaneous involvement of KS before and after treatment. Hence in patients with cutaneous KS, any

extracutaneous tumour extension including lymph nodes and subcutaneous soft tissue should show increased uptake of ^{99m}Tc-MIBI.

The assessment of cutaneous lesions on the whole body planar imaging was confined to the extremities, because the face and trunk could be partly obscured by the normal radioactive uptake of ^{99m}Tc-MIBI in the salivary glands, pharynx, myocardium, hepatobiliary, gastrointestinal, and urinary tracts (figure 8).

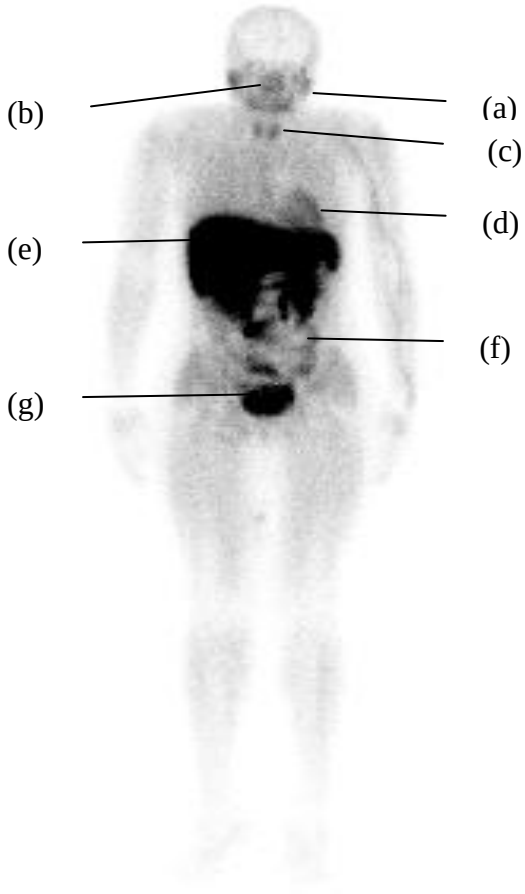


Figure 8: Anterior whole-body ^{99m}Tc-MIBI image of a 35 year old female demonstrating normal radioactive uptake in the salivary glands (a), pharynx (b), thyroid (c), myocardium (d), hepatobiliary (e), gastrointestinal (f), and urinary (g) tracts

Morphologic variants of cutaneous KS include violaceous patch, plaque, nodular, lymphadenopathic, exophytic, infiltrative, periorbital ecchymotic, telangiectatic, keloidal, and cavernous types. The macules and plaques follow skin tension lines and may disseminate rapidly and progress to form subcutaneous nodules and tumours. Histopathology of cutaneous KS includes dilated vessels and vascular slits in the dermis surrounded by spindle cells that form interlacing bundles. Erythrocytes are seen within these vascular structures interspersed between spindle cells and hemosiderin deposits. There are widespread lymphocytes, eosinophils and plasma cells in the tumour tissue (Strutton, 1991: 184; Sanders *et al*, 2004:1550).

In this study, good correlation was found between the clinical findings and detecting cutaneous KS in the extremities using ^{99m}Tc-MIBI imaging. There was higher correlation between clinical features and ^{99m}Tc-MIBI-accumulation in diffuse KS than in scattered lesions (Tables 28 & 29). Lymphatic vasculature and lymphoid tissue are prevalent in organs such as skin, gastrointestinal tract and lungs that are in direct contact with the external environment. This lymphatic distribution corresponds to the cutaneous and extracutaneous extensions of AIDS-KS. Because KS likely arises from the vasculature and lymphatic endothelium, it may present clinically with lymphadenopathy or isolated lymphoedema without cutaneous lesions (Krigel & Friedman-Kien, 1985:189). Lymphoedema may result from impaired lymphatic transport caused by KS infiltrating the lymphatic channels or lymph nodes. Lymphoedema can also be due to any of the range of OIs affecting AIDS patients.

In this study, on the clinical assessment abnormal lymph nodes were detected in only 10 patients whereas 33 patients showed abnormal lymph nodes on ^{99m}Tc-MIBI planar scans (Table 27). ^{99m}Tc-MIBI imaging provided additional information on the extent of lymph node involvement (Figures 9 and 10), which could be used for more precise staging and therapeutic planning of KS.

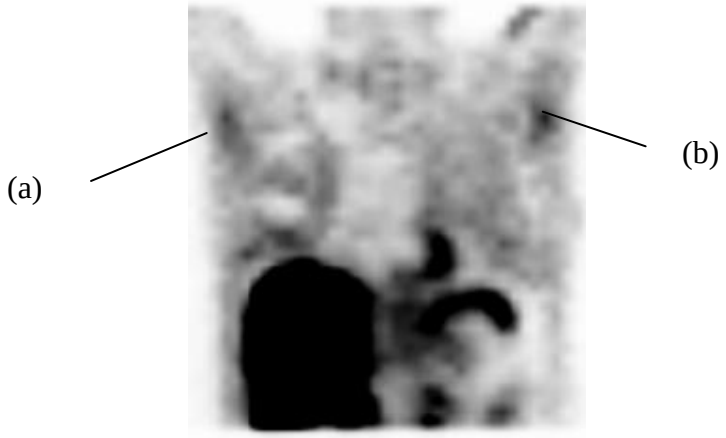


Figure 9: SPECT ^{99m}Tc-MIBI image of the chest and axilla demonstrating axillary lymph nodes (a) and (b)

Comparative studies have shown that SPECT acquisition significantly improves the sensitivity achieved with planar scintimammography when lymph nodes are small, few in number and non-palpable (Madeddu & Spanu, 2004: S23).

Discussion on SPECT imaging is confined to the chest area as only a SPECT study of the chest was performed in this study. SPECT was more effective for demonstrating abnormal lymph nodes, pericardial effusions and ascites than planar imaging. Further clinical trials to check the sensitivity of ^{99m}Tc-MIBI SPECT imaging of the abdomen and pelvis may be necessary.

Lymphoedema was also detected (figures 10 (i) and 10 (ii) in more patients on ^{99m}Tc-MIBI images than on clinical assessment (Table 26) but many false positives were present.

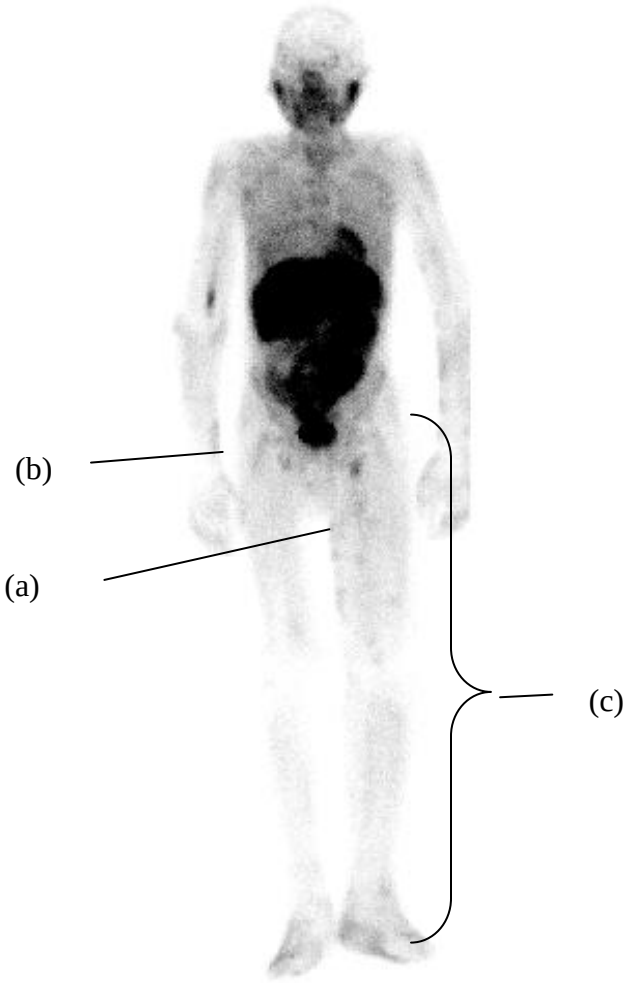


Figure 10 (i): Anterior whole-body ^{99m}Tc-MIBI image of a 34-year-old male shows radioactive uptake by the cutaneous plaques of KS in the medial left thigh (a) with inguinal lymph nodes (b) and lymphoedema of the left lower limb (c)

Lymphoedema of the lower extremity was found in four patients without any palpable inguinal lymph nodes or abnormal lymph node uptake of ^{99m}Tc-MIBI. The diagnosis of peripheral lymphoedema can be corroborated by lymphoscintigraphy.

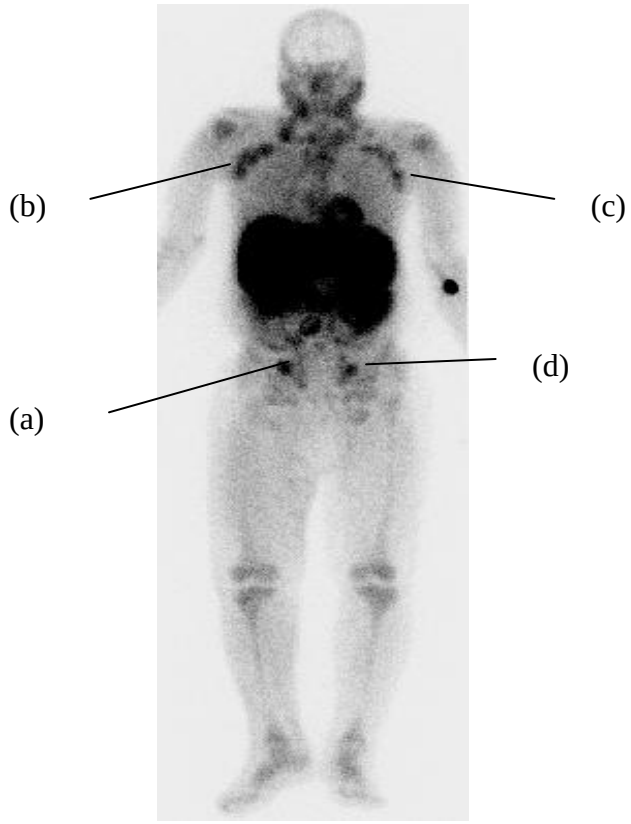


Figure 10 (ii): Anterior whole-body ^{99m}Tc-MIBI image of a 52-year-old male shows radioactive uptake by the cutaneous plaques of KS in the medial right thigh (a). Extensive lymphadenopathy is noted in multiple cervical (b), axillary (c) and inguinal lymph nodes (d) associated with lymphoedema of the right lower limb

5.3 Venous Stasis

Although in the proposal, detection of prolonged and pronounced stasis of venous activity was not mentioned as an objective, it was an incidental finding where persistent retention of radioactivity was noted in the vein proximal to the injection site of the ^{99m}Tc-MIBI scans for the duration of the study (60 minutes post injection).

Cutaneous KS is often complicated by venous stasis of the extremities, face and genitals (Krigel & Friedman-Kien, 1985:188). Clinical differential diagnosis of early KS lesions includes haemangiomas, telangiectases, stasis dermatitis and acro-angiodermatitis. Acro-angiodermatitis is related to venous insufficiency (Strutton, 1991: 184).

In this study, venous stasis was demonstrated on the ^{99m}Tc-MIBI scans proximal to the tracer injection sites, on the ipsilateral limb, in 13 patients as dilated veins with persistent retention of radioactivity at one hour (Figures 11(i) and 11(ii)). Retention of activity was seen in many more patients but these were not included in the venous stasis analysis as there was possibly some extravasation of isotope during injection. Only those images that showed no pooling of activity at the injection site were analysed. Angio-proliferative change has been found in clinically uninvolved skin around KS lesions and may account for the venous stasis demonstrated in these patients without visible cutaneous KS lesions at the injection sites (Ruszczak et al 1987: 270). The literature reviewed did not report

of such activity accumulation, hence this is possibly the first report of an imaging finding of venous stasis in KS.

(a)

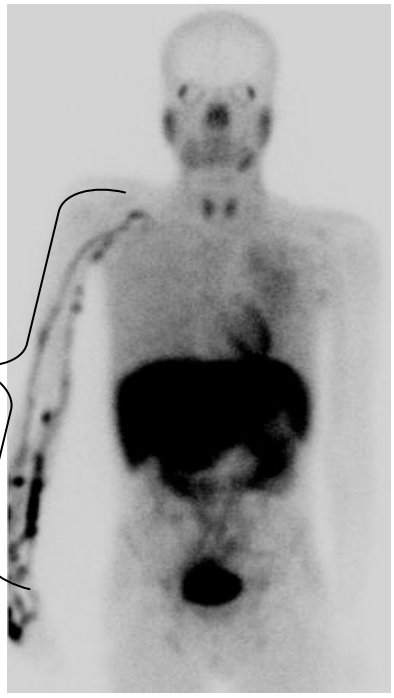


Figure 11(i): Retention of ^{99m}Tc-MIBI at 20 minutes post injection in the vein of the right arm (a) proximal to the injection site in the dorsum of the right hand

(b)

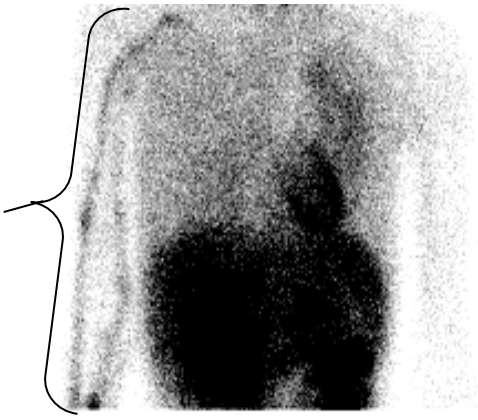


Figure 11(ii): Persistent venous stasis at 60 minutes post injection (b)

5.4 Post-treatment, Follow-up ^{99m}Tc-MIBI Scans

There are multiple treatment options for KS including systemic chemotherapy, interferon, inflammatory cytokines and treatment strategy aimed at HHV8 or angiogenesis necessary for KS growth. Patients with minimal cutaneous disease may benefit from local therapy such as excision, electron-beam or photon radiation, or intralesional chemotherapy (Sanders *et al*, 2004: 1550). The ^{99m}Tc-MIBI scans performed on patients post-treatment in this study demonstrated the therapeutic effect in patients with no uptake by skin lesions and showed decreased lymph node uptake. Low washout of ^{99m}Tc-MIBI (Table 1) is a reliable index for predicting tumour response to neoadjuvant chemotherapy whereas lack of ^{99m}Tc-MIBI-uptake in a tumour suggests resistance to some anticancer agents (Fukumoto 2004: 79). In this study, there was no or lower uptake of ^{99m}Tc-MIBI in KS of skin and lymph nodes on the follow-up scans in four patients that were imaged post- treatment (Figure12 (i) and 12(ii)). Due to the small number of patients that were imaged post treatment in this study, further studies post-treatment to assess follow-up could prove useful.

Pre-Treatment

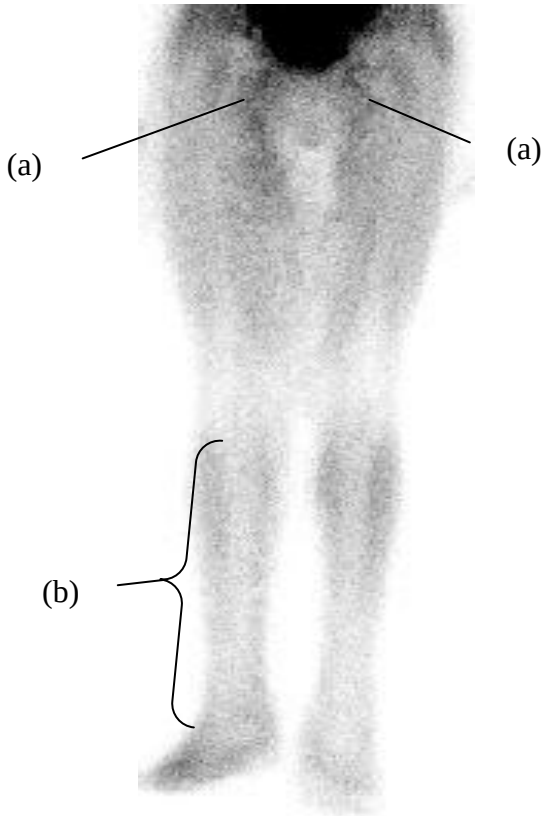


FIGURE 12(i): Pre-Treatment anterior ^{99m}Tc-MIBI scan of a 49 year old male demonstrates accumulation of radioactivity in the cutaneous plaques of KS in both upper thighs medially (a) and mild lymphoedema of the right lower extremity (b).

Post-Treatment

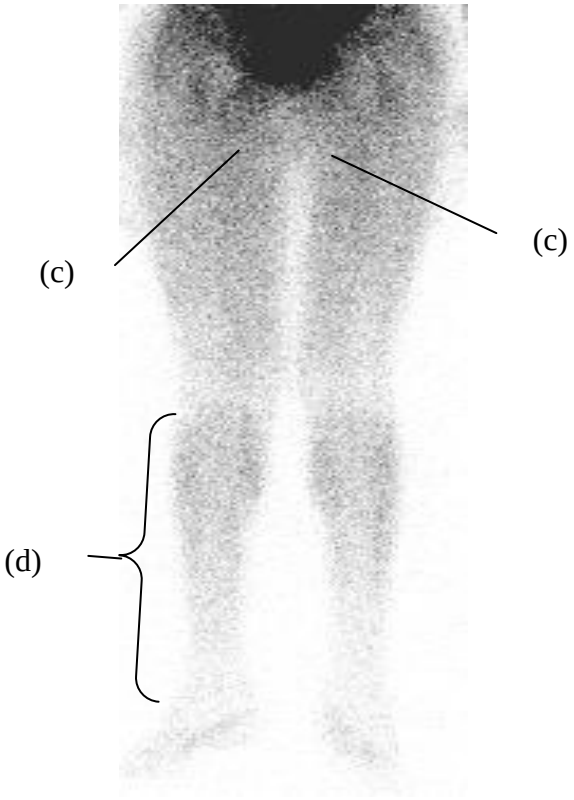


Figure 12(ii): Follow-up anterior ^{99m}Tc-MIBI scan 6 months after completion of treatment shows complete resolution of the cutaneous lesions (c) and lymphedema (d)

^{99m}Tc-MIBI was initially developed as a myocardial perfusion-imaging agent. It can be prepared readily without extra cost of the radiopharmaceutical in nuclear medicine departments that use ^{99m}Tc-MIBI for routine myocardial perfusion study. ^{99m}Tc-MIBI diffuses across plasma and mitochondrial membrane proportional to the trans-membrane potential of cells, blood flow and capillary permeability. It is localized in the mitochondria and/or attaches to a low molecular-weight protein in the lysosomes. ^{99m}Tc-MIBI accumulates in some tumours of thyroid, lung, brain, bone and breast that have higher mitochondrial density and trans-membrane electrical potential than surrounding epithelial cells (Hassan et al, 1989:337; Fukumoto, 2004: 80). Retention of ^{99m}Tc-MIBI in cells is dependent on the activity of the 170kDa P- P-gp, which is a plasma membrane protein encoded by a mammalian multidrug-resistance gene. P-gp functions as an ATP-dependent efflux pump for cytotoxic substances as well as MIBI. KS lesions of the skin and lymph nodes accumulate both ^{99m}Tc-MIBI reflecting a low expression of P-gp. Low washout of ^{99m}Tc-MIBI is a reliable index for predicting tumour response to neoadjuvant chemotherapy whereas lack of ^{99m}Tc-MIBI uptake in a tumour suggests resistance to some anticancer agents (Fukumoto 2004:79). In this study no uptake or lower uptake of ^{99m}Tc-MIBI in KS of skin and lymph nodes on the follow-up scans in patients after treatment was seen as shown in Figure 12(i) and Figure 12(ii). Hence ^{99m}Tc-MIBI imaging may be useful as a predictive test or follow-up of response of KS to treatment (Figure 13).

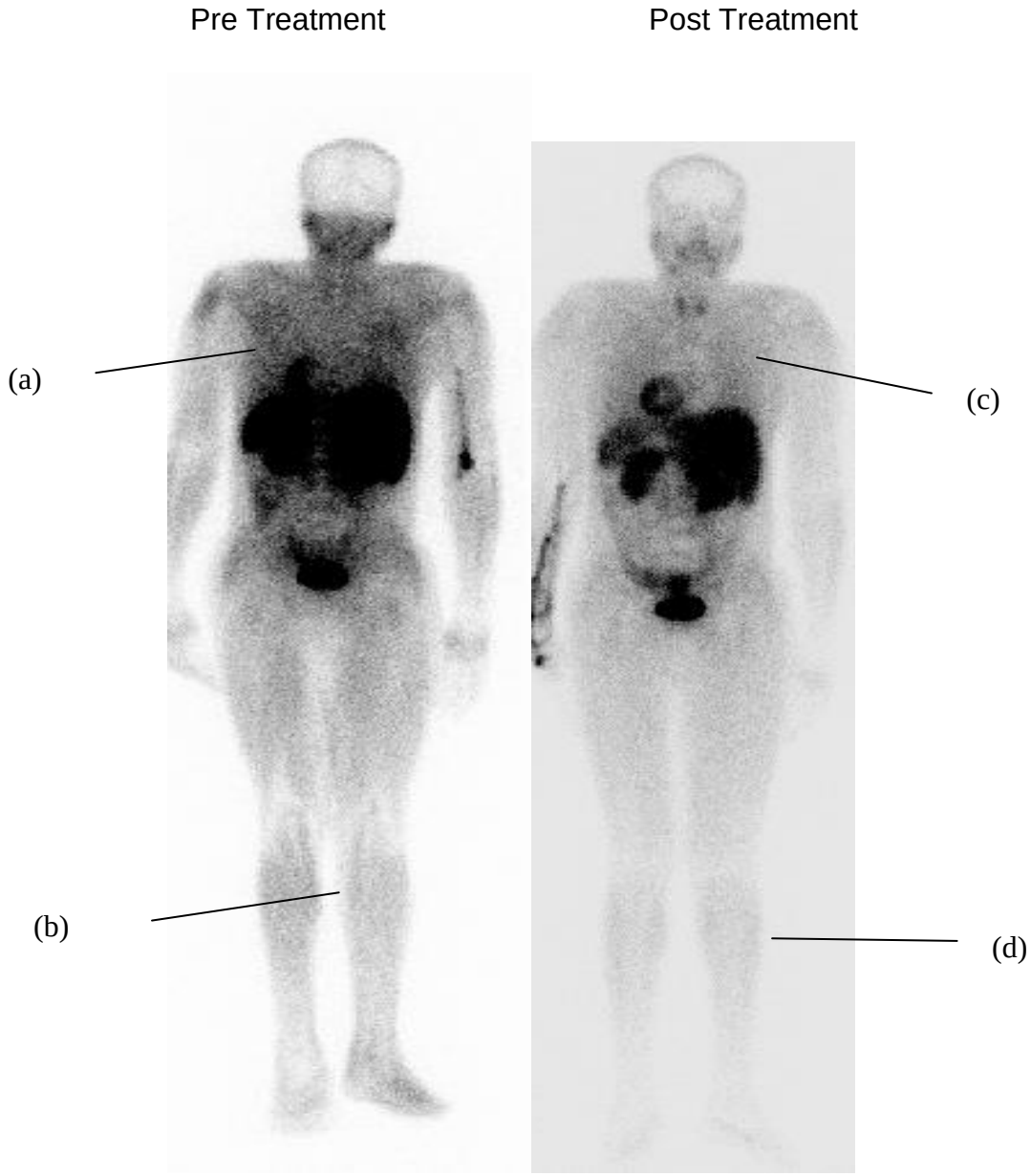


Figure 13: Pre- and Post- KS treatment ^{99m}Tc-MIBI anterior whole body images of a 36 year old male demonstrating increased radioactive uptake in the chest (a) and lymphoedema (b) of the left lower limb on the pre-treatment scan with resolution of these areas (c) and (d) on the post-treatment scan

5.5 CD4 Count Analysis

The risk of developing KS appears to decrease significantly as the patient's immune system recovers and CD4 counts increase. CD4 counts is one of the most significant prognostic factors with respect to KS, especially patients receiving highly active antiretroviral therapy should experience a decrease in KS risk if their CD4 counts increase (Mocroft *et al*, 2004: 2652).

In this study, a descriptive statistical analysis of the CD4 counts of the participants compared to their final diagnosis was made (Table 23). The analysis of the different ratios in all the groups as shown in Table 24 and in the group with KS lung involvement versus CD4 counts (Table 25) showed no statistical correlation. However it was found (Table 23), that the median of 185 for the group with KS with lung involvement was the lowest when compared to the other groups of KS with no lung involvement (median = 194) and KS with other lung pathology (median = 217). It is evident that the immune system of patients with KS lung pathology is more compromised as they have lower CD4 counts than KS patients with no KS lung involvement.

5.6 Gender Analysis

In the USA, more than 90% of KS is seen in HIV-infected men. Intrathoracic involvement is found in about one-third of men that present with cutaneous KS. Overall there is a low index of suspicion for diagnosing KS in women with AIDS possibly due to the gender bias in the differential diagnosis of KS in women with AIDS (Haramati & Wong, 2000:7414).

In this study, there was no significant difference between the distribution of the males and females within the four groups analysed (Table 15). There were 49% males and 51 % females. The groups with KS with lung involvement and KS with no lung involvement had similar sex distribution for males / females (53%/ 47% and 54%/46% respectively). The groups with KS other lung pathology and non-KS other lung pathology had identical sex distribution (40%/60% and 40%/60%).

5.7 Age Analysis

In this study, the mean age for the KS patients (n=57) was 33.6 years. The mean age of entire study population (n=72), that is, patients with AIDS-KS and AIDS Non-KS, was 36.8 years and the age of the patients ranged from 20 to 75 years. Due to one patient in the non-KS group being 75 years old, the mean age for this group was statistically significantly higher when compared to other groups (Tables 17 & 18). This was thought not to influence the overall results of the study as only one patient was affected. The ages of the patients in this study were thought to be comparable to previously published study groups. The mean age of the KS cohort that was studied by Cassol *et al* in 2004 was 31 years. In another study in Kenya on an East African population, the age-group with the highest number of KS patients was 31-40 years and an identical incidence in men and women was reported (Mwanda *et al*, 2005: 81).

CHAPTER 6

CONCLUSION

In corroboration of the hypothesis (1.3.1 and 1.3.2), it was found that ^{99m}Tc-MIBI imaging provided additional information on the extent of lymph node involvement and lymphoedema. This additional information could be used for more precise staging and therapeutic planning of KS. Follow-up scans performed on patients post-treatment demonstrated the response to KS treatment in patients with no uptake of skin lesions and also showed decreased lymph node uptake. It is evident from this study that ^{99m}Tc-MIBI imaging may be used for detecting the extracutaneous involvement of KS before and post treatment.

Although it was hypothesized that ^{99m}Tc-MIBI imaging would be able to differentiate pulmonary infection from malignancy, it was found in this study that ^{99m}Tc-MIBI imaging does not have a high sensitivity or specificity for detecting pulmonary KS. However, ^{99m}Tc-MIBI imaging could be useful for differentiating pulmonary KS in AIDS-KS patients from infection in non-KS AIDS patients.

6.1 Differentiation of Pulmonary Infection from Malignancy with ^{99m}Tc-MIBI

^{99m}Tc-MIBI imaging has a high sensitivity (97%), moderate specificity (80%) and high accuracy (92%) for differentiating pulmonary KS in AIDS-KS patients from infection in non-KS AIDS patients as shown in Table 38.

It was hypothesized that ^{99m}Tc-MIBI imaging would be able to differentiate pulmonary infection from malignancy in AIDS patients. However, as evident from Table 34, ^{99m}Tc-MIBI imaging is not ideal for differentiating pulmonary KS from

opportunistic infection in AIDS-KS patients as the sensitivity is moderate (86%), the specificity low at 50% and, accuracy at 68%.

It was evident from Table 23 that patients with pulmonary KS were the most immuno-compromised as their CD4 counts were the lowest when compared to KS patients with no KS lung involvement and non-KS patients with other lung pathology, but the difference was small.

6.2 Detection of Extracutaneous Involvement of KS using ^{99m}Tc-MIBI

It was hypothesized that ^{99m}Tc-MIBI imaging would be sensitive and specific for the detection of extracutaneous involvement of KS in AIDS patients. As is evident from Table 27, this study provided additional information on the extent of lymph node involvement. This may be useful for more accurate staging of KS and therapeutic planning therefore ^{99m}Tc-MIBI imaging may be used to demonstrate the extracutaneous involvement of KS and as a predictive test for tumour response to therapy.

SPECT compared to planar imaging with ^{99m}Tc-MIBI was more effective for demonstrating abnormal lymph nodes, pericardial effusions and ascites. Clinical trials to test the efficacy of ^{99m}Tc-MIBI SPECT imaging of the abdomen and pelvis may be considered, but the problems associated with MIBI excretion and abdominal activity would have to be addressed.

Good correlation was seen between the clinical findings and detecting cutaneous KS in the extremities using ^{99m}Tc-MIBI imaging. The correlation between the

clinical findings and ^{99m}Tc-MIBI-accummulation was better in diffuse KS than in scattered lesions (Table 28).

Lymphoedema of the lower extremity was demonstrated on the whole body planar images of patients who had no palpable inguinal lymph nodes or abnormal lymph node uptake of ^{99m}Tc-MIBI. As evident from Table 26, lymphoedema was detected in more patients on the ^{99m}Tc-MIBI images compared to the clinical assessment findings. This corroborates the hypothesis that ^{99m}Tc-MIBI imaging would be sensitive and specific for the detection of extracutaneous involvement of KS in AIDS patients.

The ^{99m}Tc-MIBI follow-up scans done on the small number of patients post-treatment demonstrated the therapeutic effect in patients with no uptake of skin lesions and showed decreased lymph node uptake as illustrated in Figure 12(i) and Figure 12(ii). In keeping with the hypothesis that ^{99m}Tc-MIBI imaging would be sensitive and specific for the detection of extracutaneous involvement after treatment in AIDS patients, it is evident from this study that ^{99m}Tc-MIBI imaging may be useful as a follow-up diagnostic tool to check the response of KS to treatment. Since only four patients had follow-up scans in this study it may be useful for statistical purposes to perform further studies with more patients to determine the response to KS treatment.

No significant difference between the distribution of the males (49%) and females (51%) was noted within the four groups analysed. The mean age for the AIDS-KS patients (n=57) was 33.6 years. The mean age of entire study population

(n=72), that is, patients with AIDS-KS and AIDS non-KS, was 36.8 years. The age of the patients ranged from 20 to 75 years.

Gender bias has been reported in the differential diagnosis of pulmonary disease in women with AIDS (Haramati & Wong, 2000:414). In this study, there was no difference in the incidence of KS according to gender.

6.3 Demonstration of Venous Stasis using ^{99m}Tc-MIBI

Venous stasis was demonstrated proximal to the injection site on the ^{99m}Tc-MIBI scans of 13 patients, as dilated veins with persistent retention of radioactivity at 60 minutes. The pathophysiological basis for this is not known. In the literature reviewed, no previously published reports of similar findings were found. Clinical trials to demonstrate venous stasis may be of value in future studies.

6.4 Concluding Remarks on the Efficacy of ^{99m}Tc-MIBI Imaging of KS in AIDS Patients

Planar whole-body ^{99m}Tc -MIBI imaging may be used as a diagnostic tool for the detection of extracutaneous involvement of KS because it provides additional information on the extent of lymph node involvement, and lymphoedema. Abnormal lymph node uptake is demonstrated better by SPECT than planar imaging. ^{99m}Tc-MIBI imaging may also be useful as a predictive test or follow-up of response of KS to treatment. These findings are novel and were not evident in the literature reviewed.

^{99m}Tc-MIBI imaging does not have high sensitivity or specificity for differentiating pulmonary KS from opportunistic infection in AIDS-KS patients.

Venous stasis was seen in dilated veins with persistent retention of radioactivity. This finding has not been described in the literature reviewed and further clinical studies to demonstrate venous stasis in AIDS-KS patients may be useful.

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APPENDICES

APPENDIX 1

i) Approval To Conduct Study - IALCH

INKOSI ALBERT LUTHULI CENTRAL HOSPITAL
PRIVATE BAG X 03
MAYVILLE
4058
TEL: (031) 240-1045 • FAX: (031) 240-1050

Date: 8 December 2003
Ref: MJ/ Research/ 99MTC

To: Mrs. F.I. Peer
Assistant Manager: Radiography
Department of Nuclear Medicine
Inkosi Albert Luthuli Central Hospital

Dear Mrs Peer,

Protocol: An evaluation of 99MTC – MIBI Imaging of Kaposi's Sarcoma in Aids Patients.

Please note that permission has been granted for you to conduct the above-mentioned study at Inkosi Albert Luthuli Central Hospital.

Yours Sincerely,

Dr M E L Joshua
Medical Manager
IALCH

ii) Approval To Conduct Study – Department of Health

01-DEC-03 12:13 TEL: P:03

PROVINCE OF
KWAZULU-NATAL
HEALTH SERVICES

ISIFUNDAZWE
SEKWAZULU-NATALI
EZEMPILO

PROVINSIE
KWAZULU-NATAL
DEPARTEMENT VAN GESONDHEID

NATALIA
330 LONGMARKET STREET
PIETERMARITZBURG

TEL. 033-3912111
FAX 033-3426744

Private Bag :X9051
Esikhwama Seposi : Pietermaritzburg
Privaatsak : 3200

Enquiries: Mr G.J. Tromp
Extension: 2761
Reference: 9/2/3/R

01 DEC 2003

Mrs F.I. Peer
P.O. Box 1435
WANDSBECK
3631

Dear Mrs Peer

REQUEST TO CONDUCT RESEARCH : AN EVALUATION OF 99M TC – MIBI IMAGING OF KAPOSI'S SARCOMA IN AIDS PATIENTS

Your letter dated 31 October 2003 addressed to Prof. Green-Thompson, refers.

Please be advised that authority is granted for you to conduct a research regarding "Evaluation 99M TC-MIBI Imaging of Kaposi's Sarcoma in AIDS Patients" provided that :-

(a) Prior approval is obtained from the Head of the relevant Institution;

(b) Confidentiality is maintained;

(c) The Department is acknowledged;

(d) The Department receives a copy of the report on completion; and

(e) The staff of the hospital are not disturbed and/or inconvenienced in their work and t patient care is not compromised.

Yours sincerely

28/11/03

SUPERINTENDENT-GENERAL
HEAD : DEPARTMENT OF HEALTH

DEPT OF HEALTH 8:44

iii) Ethics Declaration Submitted to Durban Institute of Technology

ETHICAL ISSUES CHECKLIST FOR RESEARCH APPROVAL

To be completed by all people wishing to conduct research under the auspices of Durban Institute of Technology.

1. Use the Durban Institute of Technology Research Ethics Policy and Guidelines to ensure that ethical issues have been identified and addressed in the most appropriate manner, before finalising and submitting your research proposal.

2. Please indicate [by a X as appropriate] which of the following ethical issues could impact on your research.

3. Please type the motivations/further explanations where required in the cell headed COMMENTS.

4. The highlighted response cells indicate those responses which are of particular interest to the Ethics Committee

NO.	QUESTION	YES	NO	N/A
	DECEPTION			
1.	Is deception of any kind to be used? and if so provide a motivation for acceptability.		X	
	COMMENTS:			
2.	Will the research involve the use of no-treatment or placebo control conditions? If yes, explain how subjects interests will be protected.		X	
	COMMENTS			
	CONFIDENTIALITY			
3.	Does the data collection process involve access to confidential personal data (including access to data for purposes other than this particular research project) without prior consent of subjects? If yes, motivate the necessity		X	
	COMMENTS: Informed consent to access research participant records will be obtained			
4.	Will the data be collected and disseminated in a manner that will ensure confidentiality of the data and the identity of the participants? Explain your answer	X		
	COMMENTS: The patient will be referred to in code form. The quantitative data will only be accessible to investigators of this project.			

5.	Will the materials obtained be stored and ultimately disposed of in a manner that will ensure confidentiality of the participants? If no, explain. If yes specify how long the confidential data will be retained after the study and how it will be disposed of.	X		
No.	QUESTION	**Yes**	**No**	**N/A**
	COMMENTS: The imaging data will be stored on computer during the study. After publication of the study results, the data and patient data sheet will be erased after 5 years post completion of study.			
6.	Will the research involve access to data banks that are subject to privacy legislation? If yes, specify and explain the necessity.	X		
	COMMENTS: patient records will be accessed with permission from patient, for laboratory test results and bronchoscopic results			
	RECRUITMENT			
7	Does recruitment involve direct personal approach from the researchers to the potential subjects? Explain the recruitment process	X		
	COMMENTS: The researcher on consultation with the			

	medical practitioner in charge of the patient will recruit those AIDS patients who: a. have confirmed pulmonary pathology and b. have histologically confirmed cutaneous Kaposi's sarcoma			
8	Are participants linked to the researcher in a particular relationship, for example employees, students, family? If yes, specify how.		X	
	COMMENTS			
9	If yes to 8, is there any pressure from researchers or others that might influence the potential subjects to enroll? Elaborate.			X
	COMMENTS			
10	Does recruitment involve the circulation/publication of an advertisement, circular, letter etc? Specify		X	
	COMMENTS			
11	Will subjects receive any financial or other benefits as a result of participation? If yes, explain the nature of the reward, and safeguards		X	

	COMMENTS			
12	Is the research targeting any particular ethnic or community group? If yes, motivate why it is necessary/acceptable. If you have not consulted a representative of this group, give a reason. In addition explain any consultative processes, identifying participants. Should consultation not take place, give a motivation.		X	
No	QUESTION	**Yes**	**No**	N/A
	COMMENT			
	Informed consent will be obtained			
13	Does the research fulfill the criteria for informed consent? [See guidelines]. If yes, no further answer is needed. If no, please specify how and why.	X		
	COMMENTS			
14	Does consent need to be obtained from special and vulnerable groups (see guidelines). If yes, describe the nature of the group and the procedures used to obtain permission.		X	
	COMMENTS			

15	Will a Subject Information Letter be provided and a written consent be obtained? If no, explain. If yes, attach copies to proposal. In the case of subjects who are not familiar with English (e.g it is a second language), explain what arrangements will be made to ensure comprehension of the Subject Information Letter, Informed Consent Form and other questionnaires/documents.	X		
	COMMENTS: The information letter and informed consent will be available in English and Zulu.			
16	Will results of the study be made available to those interested? If no, explain why. If yes, explain how	X		
	COMMENTS: Results will be discussed with the clinicians and published in medical journals and in the form of a dissertation. Results will be available to the patient at the end of the study.			
	RISKS TO SUBJECTS			
17	Will participants be asked to perform any acts or make statements which might be expected to cause discomfort, compromise them, diminish self esteem or cause them to experience embarrassment or regret? If yes, explain.	X		
	COMMENTS: Since the study involves AIDS patients –			

	confidentiality will be maintained.			
18	Might any aspect of your study reasonably be expected to place the participant at risk of criminal or civil liability? If yes, explain.		X	
	COMMENTS			
19	Might any aspect of your study reasonably be expected to place the participant at risk of damage to their financial standing or social standing or employability? If yes, explain.		X	
	COMMENTS			
No.	QUESTION	**Yes**	**No**	**N/A**
20	Does the protocol require any physically invasive, or potentially harmful procedures [e.g. drug administration, needle insertion, rectal probe, pharyngeal foreign body, electrical or electromagnetic stimulation, etc?] If yes, please outline below the procedures and what safety precautions will be used.	X		
	COMMENTS: A small dose of radioactivity (20 mCi ^{99m}Tc – MIBI) will be injected using an aseptic technique into a peripheral vein. This radiopharmaceutical is routinely administered for myocardial perfusion imaging and has documented safety in the medical literature. There is no			

	significant radiation or biological hazard to the patient, medical personnel or the general public. An aseptic technique will be employed during preparation of the radiopharmaceutical.			
21	Will any treatment be used with potentially unpleasant or harmful side effects? If yes, explain the nature of the side-effects and how they will be minimized.		X	
	COMMENTS			
22	Does the research involve any questions, stimuli, tasks, investigations or procedures which may be experienced by participants as stressful, anxiety producing, noxious, aversive or unpleasant during or after the research procedures? If yes, explain.		X	
	COMMENTS			
23	Will any samples of body fluid or body tissues be required specifically for the research which would not be required in the case of ordinary treatment? If yes, explain and list such procedures and techniques.		X	
	COMMENTS			
24	Are any drugs/devices to be administered? If yes, list any	X		

	drugs/devices to be used and their approved status.			
	COMMENTS: ^{99m}Tc–MIBI will be injected in a peripheral vein. The radiopharmaceutical is approved by the FDA for myocardial perfusion and tumor imaging in patients.			
	GENETIC CONSDERATIONS			
25	Will participants be fingerprinted or DNA "fingerprinted"? If yes, motivate why necessary and state how such is to be managed and controlled.		X	
	COMMENTS			
No.	QUESTION	**Yes**	**No**	**N/A**
26	Does the project involve genetic research e.g. somatic cell gene therapy, DNA techniques etc? If yes, list the procedures involved		X	
	COMMENTS			
	BENEFITS			
27	Is this research expected to benefit the subjects directly or indirectly? Explain any such benefits.	X		
	COMMENTS: Positive scan result may result in early diagnosis and appropriate treatment.			
28	Does the researcher expect to obtain any direct or indirect financial or other benefits from conducting the research? If yes, explain.	X		

	COMMENTS: The research is in fulfillment of a D Tech: Radiography degree			
	SPONSORS: INTERESTS AND INDEMNITY			
29	Will this research be undertaken on the behalf of or at the request of a pharmaceutical company, or other commercial entity or any other sponsor? If yes, identify the entity.		X	
	COMMENTS			
30	If yes to 29, will that entity undertake in writing to abide by Durban Institute of Technology Research Committees Research Ethics Policy and Guidelines? If yes, do not explain further. If no, explain.			X
	COMMENTS			
31	If yes to 30, will that entity undertake in writing to indemnify the institution and the researchers? If yes, do not explain further. If no, explain.			X
	COMMENTS			
32	Does permission need to be obtained in terms of the location of the study? If yes indicate how permission is to be obtained.	X		
	COMMENTS: The proposal and ethics approval need to be submitted to the CEO of the hospital and the Secretary General, Kwa-Zulu Natal,Dept of Health			

33	Does the researcher have indemnity cover relating to research activities? If yes, specify. If no, explain why not.	X		
	COMMENTS: The researcher has professional indemnity insurance that includes research provided it is in the scope of the practitioner. As a registered student of the Durban Institute of Technology, insurance cover is automatic.			
34	Does the researcher have any affiliation with, or financial involvement in, any organization or entity with direct or indirect interests in the subject matter or materials of this research? If yes, specify.		X	
	COMMENTS			

08-11-2001

The undersigned declare that the above questions have been answered truthfully and accurately

STUDENT NAME---------- SIGNATURE----------

DATE----------

SUPERVISOR NAME---------- SIGNATURE----------

DATE----------

APPENDIX 2

i) PARTICIPANT INFORMATION LETTER

DURBAN INSTITUTE OF TECHNOLOGY: **DEPARTMENT OF RADIOGRAPHY**

TOPIC OF RESEARCH:

^{99m} Tc- MIBI Imaging of Kaposi's Sarcoma in AIDS Patients

Dear participant

You are invited to voluntarily participate in the above project.

I am registered with the Durban Institute of Technology as a doctorate student in radiography. The aim of this study is to detect and differentiate infection from malignancy.

PROCEDURE:

A small dose of a radioactivity, will be injected into one of your veins, for example, a vein in your arm. You will then lie on the gamma camera bed for a scan. The radiation from your body will be detected by the scannner. This information is fed into a computer to produce pictures of the inside of your body. You will need another scan to check for improvement after you have completed treatment.

You are free to withdraw from this study at any time, without it affecting your treatment. Should you withdraw, it will not affect your relationship with the clinician.

RISKS / DISCOMFORT :

The amount of radiation is very small and stays in your body for only a short time.

BENEFITS:

The information obtained from this study may help in making an early diagnosis and appropriate treatment in future patients.

CONFIDENTIALITY:

All information obtained from you and from your records will be treated confidentially and will be used for research purposes only. Names will be excluded from data analysis and data presentation. Please be aware that you are free to withdraw at any stage of the project.

COST:

The study will be done free of charge with no cost to the participants.

PERSONS TO CONTACT FOR PROBLEMS OR QUESTIONS:

Fawzia Peer	Dr William Rae	Dr N Lachman
M Tech:Radiography	BSc, MB.BCh, MSc, PhD	BMed, MMed, PhD
Tel: 031 2401881	031 3272189	031 2042404
E mail: fpeer@webmail.co.za	williamrae@ialch.co.za	nirushal@dit.ac.za

ii) INFORMED CONSENT FORM

I, ……..hereby voluntarily

give consent to participate in the research entitiled:

TITLE

An Evaluation of ^{99m}Tc-MIBI Imaging of Kaposi's Sarcoma in AIDS Patients

Conducted by:

Name of researcher: Fawzia Peer

Name of supervisor: Prof Margaret Pui

Name of co- supervisors: Dr William Rae and

Dr Nirusha Lachman

PLEASE CIRCLE THE APPROPRIATE ANSWER:

1 Have you read and understood the research information sheet?

YES / NO

2 Have you had an opportunity to discuss the study?

YES / NO

3 Have you had an opportunity to ask questions regarding this study?

YES / NO

4 Have you received satisfactory answers to your questions?

YES / NO

5 Have you received enough information about the study?

YES / NO

6. Do you understand that you are free to withdraw from this study:

at any time and; without having to give reason for withdrawing?

YES / NO

7 Do you agree to voluntarily participate in this study?

YES / NO

8 Have you been counseled regarding your HIV status?

YES / NO

If you have answered NO to any of the above questions, please obtain the appropriate information BEFORE signing.

Please print clearly in block letters:

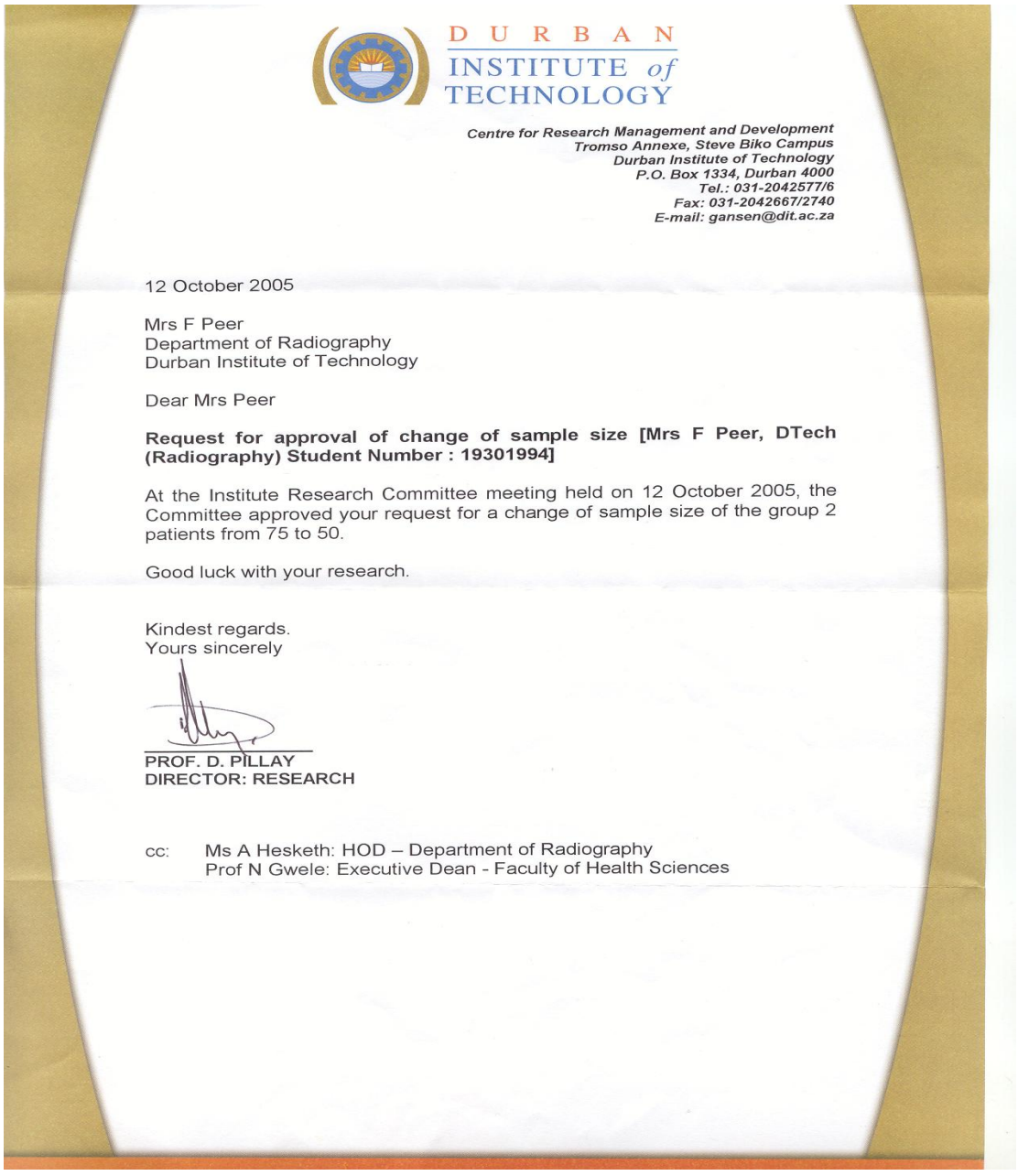
Participant Name……signature……..

Witness Name ……signature…….

Date……..

APPENDIX 3

Permission for Reduction in Sample Size



DURBAN INSTITUTE of TECHNOLOGY

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E-mail: gansen@dit.ac.za

12 October 2005

Mrs F Peer
Department of Radiography
Durban Institute of Technology

Dear Mrs Peer

Request for approval of change of sample size [Mrs F Peer, DTech (Radiography) Student Number : 19301994]

At the Institute Research Committee meeting held on 12 October 2005, the Committee approved your request for a change of sample size of the group 2 patients from 75 to 50.

Good luck with your research.

Kindest regards.
Yours sincerely

PROF. D. PILLAY
DIRECTOR: RESEARCH

cc: Ms A Hesketh: HOD – Department of Radiography
Prof N Gwele: Executive Dean - Faculty of Health Sciences

Appendix 4

Sample Spreadsheet of Results Template

ANT 10" ARM MUSCLE

Pt No	10"AntArm Area		10"AntArm counts		10"AntArm pixels		10"AntArm cts/pixel	
	PRE	POST	PRE	POST	PRE	POST	PRE	POST

ANT 10" LUNG

Pt No	10"AntLung Area		10"AntLung counts		10"AntLung pixels		10"AntLung cts/pixel	
	PRE	POST	PRE	POST	PRE	POST	PRE	POST

ANT 10" MYOCARDIUM

Pt No	10"AntMyo Area		10"AntMyo counts		10"AntMyo pixels		10"AntMyo cts/pixel	
	PRE	POST	PRE	POST	PRE	POST	PRE	POST

POST 10" ARM MUSCLE

Pt No	10"PosArm Area		10"PosArm counts		10"PosArm pixels		10"PosArm cts/pixel	
	PRE	POST	PRE	POST	PRE	POST	PRE	POST

POST 10" LUNG

Pt No	10"PosLung Area		10"PosLung counts		10"PosLung pixels		10"PosLung cts/pixel	
	PRE	POST	PRE	POST	PRE	POST	PRE	POST

POST 10" MYOCARDIUM

Pt No	10"PosMyo Area		10"PosMyo counts		10"PosMyo pixels		10"PosMyo cts/pixel	
	PRE	POST	PRE	POST	PRE	POST	PRE	POST

APPENDIX 5

Tables showing Sensitivity and 1-Specificity of data points for different chest analysis groups:

1. KS in the lung vs. other pathology (Group 1 vs. Groups 3 and 4)

Table A: Coordinates of the Curve for 60" lung/myocardium ratio to distinguish KS in lungs vs. other pathologies for Group 1 vs. Groups 3 and 4

Positive if Greater than or Equal To (a)	Sensitivity	1-Specificity
-.613886	1.000	1.000
.436218	1.000	.960
.491419	1.000	.920
.499936	1.000	.880
.513756	1.000	.840
.528363	.972	.840
.534283	.972	.800
.537003	.972	.760
.543289	.972	.720
.563623	.972	.680
.582972	.972	.640
.588638	.972	.600
.595418	.972	.560
.613932	.972	.520
.628157	.944	.520
.630256	.944	.480
.649654	.944	.440
.730037	.944	.400
.814355	.917	.400
.849397	.889	.400
.874382	.861	.400
.888357	.833	.400
.896050	.833	.360
.904497	.833	.320
.923800	.806	.320
.945831	.778	.320
.962926	.750	.320
.993521	.750	.280
1.014261	.722	.280

1.017842	.694	.280
1.052337	.694	.240
1.085383	.667	.240
1.105422	.639	.240
1.136696	.611	.240
1.148202	.583	.240
1.148839	.556	.240
1.170468	.528	.240
1.194349	.500	.240
1.201723	.500	.200
1.217535	.472	.200
1.231562	.444	.200
1.235200	.417	.200
1.251006	.389	.200
1.266127	.361	.200
1.275420	.361	.160
1.311009	.333	.160
1.361724	.306	.160
1.435460	.306	.120
1.494282	.278	.120
1.509344	.250	.120
1.524776	.222	.120
1.542388	.222	.080
1.579357	.194	.080
1.617808	.167	.080
1.627730	.139	.080
1.646973	.111	.080
1.869202	.111	.040
2.089487	.083	.040
2.113038	.083	.000
2.175729	.056	.000
2.753790	.028	.000
4.276086	.000	.000

a: The smallest cutoff value is the minimum observed test value minus 1, and the largest cutoff value is the maximum observed test value plus 1. All the other cutoff values are the averages of two consecutive ordered observed test values.

2. KS in lung vs. all others (Group 1 vs. Groups 2,3 and 4)

Table B: Coordinates of the Curve for 60" lung/myocardium ratio to distinguish KS in lungs vs. all others for Group 1 vs. Groups 2,3 and 4

Positive if Greater than or Equal To (a)	Sensitivity	1 - Specificity
-.613886	1.000	1.000
.436218	1.000	.972
.491419	1.000	.944
.499936	1.000	.917
.513756	1.000	.889
.528363	.972	.889
.534283	.972	.861
.537003	.972	.833
.543289	.972	.806
.563623	.972	.778
.582972	.972	.750
.588638	.972	.722
.595418	.972	.694
.613932	.972	.667
.628157	.944	.667
.630256	.944	.639
.649654	.944	.611
.730037	.944	.583
.806967	.917	.583
.829320	.917	.556
.838729	.889	.556
.851418	.889	.528
.864896	.861	.528
.877192	.861	.500
.888357	.833	.500
.896050	.833	.472
.904497	.833	.444
.921580	.806	.444
.938451	.806	.417
.945831	.778	.417
.962926	.750	.417
.993521	.750	.389
1.014261	.722	.389
1.017842	.694	.389
1.031664	.694	.361
1.044842	.694	.333

1.065514	.694	.306
1.085383	.667	.306
1.105422	.639	.306
1.136696	.611	.306
1.148202	.583	.306
1.148839	.556	.306
1.170468	.528	.306
1.192194	.500	.306
1.194860	.500	.278
1.201723	.500	.250
1.217535	.472	.250
1.231562	.444	.250
1.235200	.417	.250
1.251006	.389	.250
1.266127	.361	.250
1.275420	.361	.222
1.311009	.333	.222
1.361724	.306	.222
1.400380	.306	.194
1.449726	.306	.167
1.494282	.278	.167
1.509344	.250	.167
1.524776	.222	.167
1.542388	.222	.139
1.553987	.194	.139
1.583187	.194	.111
1.617808	.167	.111
1.627730	.139	.111
1.646973	.111	.111
1.802623	.111	.083
2.006281	.111	.056
2.089487	.083	.056
2.113038	.083	.028
2.133381	.056	.028
2.189146	.056	.000
2.753790	.028	.000
4.276086	.000	.000

a: The smallest cutoff value is the minimum observed test value minus 1, and the largest cutoff value is the maximum observed test value plus 1. All the other cutoff values are the averages of two consecutive ordered observed test values.

3. KS vs. Non-Ks other pathology (Group 1, 2 and 3 vs. Group 4)

Table C: Coordinates of the Curve for 60" lung/myocardium ratio to distinguish KS from non KS for Group 1, 2 and 3 vs. Group 4

Positive if Greater than or Equal To (a)	Sensitivity	1 - Specificity
-.613886	1.000	1.000
.436218	1.000	.933
.491419	1.000	.867
.499936	1.000	.800
.513756	1.000	.733
.528363	.982	.733
.534283	.982	.667
.537003	.982	.600
.543289	.982	.533
.563623	.982	.467
.582972	.982	.400
.588638	.982	.333
.595418	.982	.267
.613932	.982	.200
.628157	.965	.200
.630256	.965	.133
.649654	.965	.067
.730037	.965	.000
.806967	.947	.000
.829320	.930	.000
.838729	.912	.000
.851418	.895	.000
.864896	.877	.000
.877192	.860	.000
.888357	.842	.000
.896050	.825	.000
.904497	.807	.000
.921580	.789	.000
.938451	.772	.000
.945831	.754	.000
.962926	.737	.000
.993521	.719	.000
1.014261	.702	.000
1.017842	.684	.000
1.031664	.667	.000
1.044842	.649	.000
1.065514	.632	.000

1.085383	.614	.000
1.105422	.596	.000
1.136696	.579	.000
1.148202	.561	.000
1.148839	.544	.000
1.170468	.526	.000
1.192194	.509	.000
1.194860	.491	.000
1.201723	.474	.000
1.217535	.456	.000
1.231562	.439	.000
1.235200	.421	.000
1.251006	.404	.000
1.266127	.386	.000
1.275420	.368	.000
1.311009	.351	.000
1.361724	.333	.000
1.400380	.316	.000
1.449726	.298	.000
1.494282	.281	.000
1.509344	.263	.000
1.524776	.246	.000
1.542388	.228	.000
1.553987	.211	.000
1.583187	.193	.000
1.617808	.175	.000
1.627730	.158	.000
1.646973	.140	.000
1.802623	.123	.000
2.006281	.105	.000
2.089487	.088	.000
2.113038	.070	.000
2.133381	.053	.000
2.189146	.035	.000
2.753790	.018	.000
4.276086	.000	.000

a: The smallest cutoff value is the minimum observed test value minus 1, and the largest cutoff value is the maximum observed test value plus 1. All the other cutoff values are the averages of two consecutive ordered observed test values.

4. **KS in lungs vs. Non-Ks other pathology (Group 1 vs. Group 4)**

Table D: Coordinates of the Curve for 60" lung/myocardium ratio to distinguish KS in the lungs from non KS for Group 1 vs. Group 4

Positive if Greater than or Equal To (a)	Sensitivity	1 - Specificity
-.613886	1.000	1.000
.436218	1.000	.933
.491419	1.000	.867
.499936	1.000	.800
.513756	1.000	.733
.528363	.972	.733
.534283	.972	.667
.537003	.972	.600
.543289	.972	.533
.563623	.972	.467
.582972	.972	.400
.588638	.972	.333
.595418	.972	.267
.613932	.972	.200
.628157	.944	.200
.630256	.944	.133
.649654	.944	.067
.730037	.944	.000
.814355	.917	.000
.849397	.889	.000
.874382	.861	.000
.896804	.833	.000
.923800	.806	.000
.945831	.778	.000
.981584	.750	.000
1.014261	.722	.000
1.050839	.694	.000
1.085383	.667	.000
1.105422	.639	.000
1.136696	.611	.000
1.148202	.583	.000
1.148839	.556	.000
1.170468	.528	.000
1.199058	.500	.000
1.217535	.472	.000
1.231562	.444	.000

1.235200	.417	.000
1.251006	.389	.000
1.275390	.361	.000
1.311009	.333	.000
1.411071	.306	.000
1.494282	.278	.000
1.509344	.250	.000
1.532544	.222	.000
1.579357	.194	.000
1.617808	.167	.000
1.627730	.139	.000
1.850631	.111	.000
2.096412	.083	.000
2.175729	.056	.000
2.753790	.028	.000
4.276086	.000	.000

a: The smallest cutoff value is the minimum observed test value minus 1, and the largest cutoff value is the maximum observed test value plus 1. All the other cutoff values are the averages of two consecutive ordered observed test values.