REFERENCE VALUES FOR THYROID UPTAKE OF TECHNETIUM-99m PERTECHNETATE FOR THE NAMIBIAN POPULATION

By

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Dissertation submitted in fulfilment of the requirements for the degree Master of Technology: Radiography (Nuclear Medicine)

In the

Faculty of Health and Wellness Sciences

At the

CAPE PENINSULA UNIVERSITY OF TECHNOLOGY

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Date submitted (October 2012)

DECLARATION

I, **Roswita Hambeleleni Hamunyela,** declare that the contents of this dissertation/thesis represent my own unaided work, and that the dissertation/thesis has not previously been submitted for academic examination towards any qualification. Furthermore, it represents my own opinions and not necessarily those of the Cape Peninsula University of Technology.

Signed **Date**

SYNOPSIS

Thyroid physiology and structure can be evaluated by scintigraphic imaging. Iodine-131, discovered in the late thirties was the first radioisotope to be used for thyroid uptake assessment, since then thyroid uptake and imaging continues to play a vital role in different thyroid related clinical situations. Because Iodine-131 has serious disadvantages related to high radiation dose, it has been limited to staging and follow up of patients with thyroid carcinomas. Its substitute Iodine -123 has proved to be suitable in terms of shorter half-life and better imaging energy. Technetium-99m pertechnetate, because of its availability is also used for thyroid imaging and uptake assessment. The similarity of the Technetium-99m pertechnetate ions and iodide is the explanation for the thyroid glands ability to absorb it.

Historically, chronological changes in the normal values of thyroid uptake normal values have been reported in different geographical areas. These changes have been linked to geographical and chronological fluctuations in dietary iodine intake in different populations. Technetium-99m pertechnetate, Iodine-131 and Iodine-123, all reflect thyroid iodide accumulation. In areas where there is iodine deprivation there will be elevated radioisotope uptake in euthyroid persons, and radiotracer uptake is even higher in areas of Iodine deficiency. Conversely in areas where a population is exposed to stable iodine and abundant amount of iodine intake from other sources, radiotracer uptake will be decreased.

Namibia is a country with mixed ethnicity consisting mainly of Africans (Hereros, Ovahimba, Kavangos, Caprivians, Ovambo, Damaras, Namas, Bushmen, Coloureds and Caucasians). The population has access to dietary iodine and all ethnic groups consume reasonable amounts of food containing iodine such as bread, dairy products, table salt and sea food.

Similar to other Nuclear Medicine departments worldwide, Namibia makes use of Technetium-99m pertechnetate for thyroid uptake and imaging. However thyroid uptake quantification needs to be related to normal values as a point of reference. Despite reports of thyroid uptake fluctuations of normal values and reports emphasising the importance of periodic checks on the thyroid uptake normal values, Namibia has never determined these reference values.

This study examines the fundamental statement posed in the hypothesis that the thyroid uptake reference values for Technetium-99m pertechnetate in a Namibian population deviate from available International normal reference values.

Eighty three participants considered to be euthyroid from Windhoek, Namibia received Technetium-99 m pertechnetate as part of their evaluation. The euthyroid state was based on a combined evaluation of clinical history, palpation of the thyroid gland and assessment of thyroid hormones (TSH, T3 and T4).

The objectives of the study were:

- To establish normal reference values of Technetium-99m pertechnetate uptake in euthyroid persons in Namibia.
- To compare the outcome reference values to current available existing International normal reference values.
- To determine possible factors contributing to the deviation of thyroid uptake reference values in the Namibian population.
- To recommend reference values for Namibia.

This was achieved by studying the unmedicated 83 participants with their iodine diet, rather than patients with thyroid or other diseases. The participants reported to the Nuclear Medicine department. The participants completed a questionnaire designed to reveal the presence of thyroid pathologies or any other pathology. The completed questionnaire was reviewed and if the participant fulfilled the requirements for the study, blood was drawn for thyroid hormone assessment. The participants were then given 100MBq Technetium-99m pertechnetate intravenously. Using a low energy high resolution dual head gamma camera, the thyroid uptake was recorded at 20 minutes.

There was a marked difference between the International normal values (0.75% to 4%) used in the Nuclear Medicine department and those found in this study. The results of seven subjects were excluded due to abnormal blood results, and technical errors, bringing the actual sample size to 76 euthyroid participants. The age range was 39 to 81 years, and there were 58 females and 18 males. The mean uptake for the euthyroid group was found to be 0.78%± 0.45%.

The Kolmogorov-SmirnovaLilliefors Significance parametric test presented us with results that state that our population has a non-normal distribution, and therefore the standard deviation should not be applied to determine the normal values. The statistical test for skewness and kurtosis was adopted in this study to correct for skewness and the normal values determined were 0.35% to 1.22% (95.5% CI) of Technetium-99m pertechnetate at 20 minutes. An alternative method used to determine the normal reference range for Technetium-99m pertechnetate in a study sample obtained from a non- normal distribution was determined using the 5^{th} and the 95^{th} percentiles .The normal reference range using the 5^{th} and the 95^{th} percentile was 0.17% - 1.7%.

This study provides new evidence supporting the importance of periodic checks on normal reference values. The evidence is provided by the analysis of the empirical data obtained in this study of a population that has sufficient daily intake of stable iodine. The results obtained

from this study and other studies proving the difference in thyroid uptake between different populations and geographical regions makes it incumbent upon laboratories to establish a normal reference range for a particular geographical location. Furthermore the study attests to the importance of re-evaluating normal values for thyroid uptake as part of a quality assurance programme.

ACKNOWLEDGEMENTS

I wish to thank:

- Mrs G Philotheou (Supervisor) for her professional guidance, her time and dedication in making this research a success.
- Dr T Kotze (Co-Supervisor) for her engagement as a Nuclear Medicine Physician in providing scientific advice.
- Dr S C Herman for being available at the research site as a Nuclear Medicine Physician providing technical advice in the data collection process.
- Veliswa Tshetsha (CPUT Tygerberg Hospital Campus Library), for always being available to teach me literature searching methods.
- Ms Magdalena H Owoses (Nuclear medicine Technologist) and Ms M Mandume (Nuclear Medicine Nurse), not just research assistants, but best friend and mother figure in my life, thank you for assisting in the data collection process.
- The Department of Nuclear Medicine, Windhoek Central Hospital, thank you for permitting me to conduct my research in the department.
- The Namibian Institute of Pathology, for testing the serum thyroid hormone values on the research subjects.
- Radiation Protection Department, Ministry of Health, for providing radiation ethical advice.
- All the research participants, thank you for coming forth to take part in this research that will help us better manage our patients with thyroid disorders in Namibia.

The financial assistance of the Namibian Government Scholarship and Training Program (NGSTP) and the Ministry of Health and Social Services (Namibia) towards this research is acknowledged. Opinions expressed in this dissertation and the conclusions arrived at, are those of the author, and are not necessarily to be attributed to the NGSTP or the Ministry of Health.

DEDICATION

This thesis is dedicated to my family

My father Mr Jason K Hamunyela, my mother Mrs RoswitaNHamunyela,

Thank you for your constant love and support and always believing in me.

My siblings Thomas ***"Tommy"*** Hamunyela, David N***"NAD"*** Hamunyela,

Kondjela S ***"K"*** Hamunyela, Kwathiindje ***"Kwathi"*** Hamunyela

And Inotila M ***"Ino"*** Hamunyela.

May you be inspired to always be great achievers.

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ABREVIATIONS

ACR	American College of Radiology
IAEA	International Atomic Energy Agency
ICRP	International Commission of Radiological Protection
MRC	Medical Research Council –South Africa
SNM	Society of Nuclear Medicine (International)
WHO	World Health Organisation
ROI	Region of Interest

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1. CHAPTER ONE INTRODUCTION

The normal reference values for thyroid uptake of Technetium-99m pertechnetate, or radioactive iodine in euthyroid persons changes with the geographic location over time Nelson et al (1970).
The important contribution of thyroid uptake in the evaluation of the thyroid gland was the motivation for studies to determine normal reference values for thyroid uptake, as the comparison of thyroid uptake to these normal reference values are what determine the presence of a thyroid disorders.

Throughout history fluctuations in the normal thyroidal reference range have been noted. Bernard et al (1970) discovered that there was a deviation from traditional normal ranges that they used routinely and the range found in their normal subjects; furthermore Bernard et al (1970) stated that a wide range of normal radioiodine uptake values made it incumbent on the Nuclear Medicine facility to establish a normal range for the specific geographical area. Bernard et al (1970) conclude that the iodine uptake ranges have definitely changed over the years, partly due to variations in iodine intake of a population, and that all Nuclear Medicine facilities should establish ranges for iodine uptake studies as necessary with time change and equipment. In California, Blum and Chandra (1971) likewise reported that several laboratories had recorded a lowering of radioactive iodine intake by the normal human thyroid gland. Similarly in Minneapolis, Wong and Schultz (1977) reported that the radioiodine normal ranges had deviated twice in a span of 20 years.
More so in a relatively stable population in Ft. Smith, Arakansas, Culp and Huskison (1978) also reported that there was a deviation in the normal uptake values of Iodine -131 (^{131}I). In Boston, Anderson and Powsner (1996) observed stability in the normal values of radioactive iodine in the previous 16 years. Although there were no significant changes in values for thyroid uptake of Iodine -123 (^{123}I) in this study; Anderson and Powsner (1996) advise that thyroid uptake results should be monitored periodically to maintain quality assurance for thyroid uptake studies.

Namibia has a population of 1.8 million, with mixed ethnicity consisting mainly of Africans (Hereros, Ovahimbas, Kavangos, Caprivians, Ovambo, Damaras, Namas, Bushmen, and Coloureds) and Caucasians. About 50% of the population constitutes the Ovambo tribe other ethnic groups include 9% Kavango, 7% Herero, 7% Damara, 5% Nama, 4% Caprivian, 3% Bushmen, 2% Coloureds and 0.5% Tswana (Namibia National Planning Commission, 2011).

The country's geographical location consists of deserts and vast arid regions, bushveld and swamps. In Namibia the reference range used for thyroid uptake of Technetium-99m pertechnetate is 0.75% to 4 % of the administered dose at 20 minutes. This range has been adapted from international Nuclear Medicine guidelines, as it is not always convenient for each Nuclear Medicine facility to determine Technetium-99m pertechnetate uptake in normal euthyroid individuals. There has been no specific reference range worked out for this geographical region.

The present study sought to investigate the fundamental statement in the research hypothesis; *"Thyroid uptake reference values for Technetium-99m pertechnetate in a Namibian population deviate from available International normal reference values".* This study further sought to establish normal reference values of Technetium-99m pertechnetate uptake in euthyroid persons for the Namibian population and compare the outcome reference values to current available existing international normal reference values used for the Namibian population.

1.1 Rationale

The thyroid uptake reference range has been used extensively in Nuclear Medicine to evaluate patients with suspected thyroid disorders. The normal reference range is obtained by taking a large group of people (O'Brien et al (1983), suggests a sample of 100 values) in a population to represent that population, then to measure their thyroid uptake values and calculate the mean.

Normal reference values for radioactive iodine uptake, are obtained from historical data and range from 5% - 15% at 4 hours and 10% - 35% at 24 hours. Technetium-99m pertechnetate uptake in the thyroid gland is estimated to be 1.5% - 2% of the injected activity within 20 min (Canon, 2010). The reference values for the thyroid uptake test used in Namibia are 0.75% - 4 % uptake of Technetium-99m pertechnetate at 20 min. These values have been adopted from the international guidelines (American College of Radiology (ACR) - Society of Nuclear Medicine (SNM) – Society of Paediatric Radiology (SPR), 2006).
The historical reference values should not be strictly used since uptake may vary depending on equipment used in different institutions and specific groups with varying levels of dietary iodine intake in different geographical regions (Canon, 2010).

This study sought to determine reference values for thyroid uptake of Technetium-99m pertechnetate for the Namibian population in order to review the current normal values being used in the department, and in turn improve patient management.

1.2 Problem Statement

There are existing international Nuclear Medicine normal reference values (ACR-SNM-SPR, 2009 and SNM, 2006) for thyroid uptake studies using Technetium-99m pertechnetate. These have been established by measuring Technetium-99m pertechnetate uptake in the thyroid gland of euthyroid persons. Previous research has found that the thyroid uptake normal values vary from one geographical location to another and from one decade to another Nelson et al (1970). Deviations in these values are primarily linked to the fact that the level of dietary iodine intake differs from one population to another. There has been no thyroid uptake reference values worked out for the Namibian population.

1.3 Research Question

Do the normal reference values for thyroid uptake of Technetium-99m pertechnetate in a Namibian population differ from current traditionally used international reference values?

1.4 Significance of the Study

This study on thyroid uptake reference values sought to determine the normal reference values for the Namibian population. Should the reference values obtained in this study be adopted by the Nuclear Medicine department of the Windhoek Central Hospital, they will improve the accuracy /validity of the thyroid uptake investigation. Furthermore these values will afford better management of patients referred to the department with suspected thyroid disorders.

1.5 Delimitations

This research study determined the normal values of Technetium-99m pertechnetate uptake in normal euthyroid participants except pregnant and breast feeding females from 40 years of age as well as patients with normal thyroid functions who have already been referred for a thyroid scan in a representative sample of the Namibian population in Windhoek between

September 2011 and December 2011. The study took place at the only state Nuclear Medicine department in Namibia at the time; Windhoek Central Hospital, which had the appropriate dual head gamma camera and Low Energy High Resolution collimator.

1.6 Research Aim and Objectives

The overall aim of the study was to establish the normal reference values for thyroid uptake of Technetium-99m pertechnetate in the Namibian population by studying unmedicated euthyroid subjects from the Namibian population rather than patients with thyroid disease and or other diseases.

The objectives of the studies were as follows:

1. To compute normal reference values of Technetium-99m pertechnetate uptake in euthyroid persons in Namibia.

2. To compare the outcome reference values to current available existing international normal reference values.

1.7 Definition of Terms

- **^{131}I, ^{123}I, ^{125}I,**

Various isotopes of iodine used in the nuclear medicine clinical context.

- **Technetium-99m pertechnetate (^{99m}Tc-pertechnetate)**

A gamma-emitting radionuclide imaging agent used for the diagnosis of disease in many human body tissues.

- **T1/2 or half life**

The mean time needed for half the nuclei in a radioactive sample substance to undergo radioactive decay.

- **keV**

A unit of energy (kiloelectron volt) equal to the energy gained by an electron in being accelerated through the potential difference of 1 volt (one thousand electron volts)

- **MBq**

Mega Bequerel is the International System of Units (SI unit) for radioactivity; it is defined as the activity of radioactive material in which a nucleus decays per second.

- **mCi**

milli Curie a non-SI unit of radioactivity equating to $3.7x10^{7}$disintegrations per second

- **Thyroid uptake**

Thyroid uptake is a measurement that involves the use of radionuclide and a radioactive detector (gamma camera or gamma probe) to assess the function of the thyroid gland.

- **Normal reference range**

The range in concentration of particular substances found in normal healthy tissues and secretions

- **Euthyroid**

The state of having normal thyroid gland function

- **^{99}Mo- ^{99m}Tc generator**

A Technetium-99m generator, or a Technetium ‘cow’ or ‘Moly cow’, is a generator used to elute the metastable isotope Technetium from a source of decaying Molybdenum-99.

- **TSH, T3, T4**

T3 and T4 are thyroid hormones. The TSH is thyroid-stimulating hormone and it is secreted by the pituitary gland after a signal from the hypothalamus through another hormone called TRH or thyrotropin releasing hormone.

- **PBI**

Protein-bound Iodine is thyroid serum hormone in its circulating form, comprising of one or more iodothyronines coupled to one or more of the serum proteins.

1.8 Overview of Dissertation Structure

In order to understand the process of establishing the normal reference values for thyroid uptake of Technetium-99m pertechnetate for the Namibian population the succeeding chapters delineate the areas of research and theory that inform the stated research question.

Chapter 2 Literature review

This chapter reviews the literature relevant to the current study, analysis critically and summarises similar studies done to establish the normal reference values for thyroid uptake either with radioactive Iodine or Technetium-99m pertechnetate.

Chapter 3 Research Methodology

The research methodology in a systematic manner describes the data collection process; furthermore it denotes the rationality for selecting the different research methods and procedures.

Chapter 4 Research Findings

This chapter on research findings presents the overall research determinations of the present study.

Chapter 5 Discussion

The discussion chapter discourses the overall research findings of the study. It further compares the recent findings to existing literature on thyroid uptake reference values.

Chapter 6 Conclusion

This chapter resolves the research on establishing normal reference values for thyroid uptake of Technetium-99m pertechnetate for the Namibian population.

2 CHAPTER TWO REVIEW OF RELATED LITERATURE

The thyroidal uptake of Technetium-99m pertechnetate is a widely used clinical index of the thyroid organs functional state. Since the thyroid uptake test is frequently used, there is a danger of established normal reference values continuing to be used long after they should have been revised.

Historically Nuclear Medicine physicians have stated that the reference values need to be determined for different Nuclear Medicine facilities. Since the 1950's different Nuclear Medicine facilities have been establishing their own normal reference values Anderson et al (1996).In more recent literature, Elgazzer (2011) concedes that normal reference ranges are different for different populations, as well as differing instrumentation used. Thus normal reference values need to be determined for each Nuclear Medicine facility.

This chapter refers to previous research relevant to this study either theoretically or empirically. The purpose of this chapter is to locate this study in the context of what is already known about thyroid uptake reference values, and makes a case for the necessity to review the thyroid uptake reference values. The literature review identifies the key variables to consider when establishing these values, highlights the existing theories on deviations in the reference values and the different problems, limitations and inconsistencies in the existing research.
Furthermore to answer the research question *"Do the normal reference values for thyroid uptake of Technetium-99m pertechnetate in a Namibian population differ from current International reference values?"* the following areas will be reviewed : similarities or differences in terms of research aims/objectives, research design and sampling, radiopharmaceuticals, serum thyroid hormones, anatomy and physiology, pharmacokinetics of radiopharmaceuticals ,instrumentation and imaging protocols, thyroid uptake calculations, the different methods of data analysis, and finally the results and findings of the different studies.

At the end of this chapter it will be clear how the existing research relates to this study, the importance of reviewing thyroid uptake reference values and the possible contribution the present study will make to the population being studied.

2.1 Thyroid Disorders in Namibia

In Namibia approximately 21 cases of thyroid cancer are diagnosed each year. There are no endocrinologists in Namibia. A typical diagnostic work-up consists of clinical assessment, biochemical thyroid function tests and a Technetium-99m pertechnetate thyroid scan. The patient is then referred to a surgeon who performs the appropriate operation, usually near-total thyroidectomy. When pathology confirms the diagnosis, the patient is seen by a medical oncologist and dispatched to South Africa to a radiation oncology department for ^{131}I whole body scan and ^{131}I therapy. The legal limit of a single ^{131}I dose that can be administered as an outpatient is 555 MBq (15 mCi). This is the maximum dose used to treat hyperthyroidism in Namibia and much lower than an ablative dose for thyroid cancer. Since all thyroid cancer therapy is in South Africa, follow-up of post ^{131}I therapy patients treated for thyroid cancer is also performed in South Africa. However, occasionally ^{131}I Whole Body Scan (WBS) is performed using an administered dose of 37 MBq (1 mCi). Serum thyroglobulin measurement, FDG PET imaging and ^{123}I imaging are not available in Namibia. However, ^{99m}Tc Sestamibi and ^{201}Tl WBS can be performed to determine the presence of thyroid cancers (IAEA publication, 2009).

2.2 Thyroid Uptake Normal Values

The usefulness of the thyroid uptake test in part depends on an established range of values for the test in a normal healthy euthyroid population.
Canon (2010) gives the normal reference values for radioactive iodine uptake as 5% - 15% at 4 hours and 10% - 35% at 24 hours, and the values for Technetium-99m pertechnetate uptake as 1.5% - 2% of the injected activity within 20 min, while Ramos et al, 2002 states that the values for Technetium-99m pertechnetate uptake are 0.3% - 3 % at 20 minutes .Furthermore Canon (2010), advises that the historical reference values should not be strictly used since uptake may vary depending on instrumentation used in different institutions and specific groups with varying levels of dietary iodine intake in different geographical regions. The previous literature presents similarities in research aims/objectives. The main aim of reviewing existing literature was to determine thyroid uptake normal values, due to reported fluctuations of the normal thyroidal radioiodine uptake from widely separate geographic locations. Though the distinct aim in all previous literature was to establish the normal values, there were additional issues to overall objectives of some studies which made them slightly different from each other. Al-Muqbel and Tashtoush (2010) while determining values for the Jordan population concurrently added determining values for patients with

Hashimotos thyroiditis as well as early phase subacute thyroiditis. Milakovic et al (2006) added patients with thyroid nodularity and compared the euthyroid group with those of thyrotoxic individuals and observations from previous literature. Sachs et al (1972) and Blum and Chandra (1971) were more creative in their research, and went to the extent of investigating the amount of iodine in the commercially baked bread in New York and compared this to the iodine in bread from Columbia and Missouri.

The distinct aim of the present study seeks to determine reference values for thyroid uptake of Technetium-99m pertechnetate for the Namibian population as there is a lack of standard values for Technetium-99m pertechnetate uptake by the thyroid gland.

2.3 Thyroid Uptake and Scintigraphy

The thyroid uptake measurement is a common procedure performed in Nuclear Medicine departments. It is a measure of the amount of radioactive iodine or Technetium-99m pertechnetate that accumulates in the thyroid at different times depending on the radiotracer being used (ARC, SNM, SPR revised guidelines, 2009).

2.3.1 Thyroid Uptake Clinical Indications

There are several definite indications for the thyroid uptake procedure including but not limited to the following:

1. Making a distinction between hyperthyroidism and other types of thyrotoxicosis (ARC, SNM, SPR revised guidelines, 2009). Hyperthyroidism or otherwise thyrotoxicosis as defined by Christian and Waterstram-Rich (2012) as a thyroid disorder that is associated with an increased body metabolism and the overall effect of raised metabolic activity throughout the whole body. Furthermore hyperthyroidism causes increased thyroid hormone levels in the blood circulation, which in turn is caused by thyroid hormones being produced and released in large quantities by the thyroid gland. The aetiology of this thyroid disorder is Graves' disease

2. To determine the dose of ^{131}I for the treatment of hyperthyroidism and the ablative dose for therapy (ARC, SNM, SPR revised guidelines, 2009).

To establish the normal reference values of a population the thyroid uptake procedure done routinely in Nuclear Medicine facilities is used to obtain uptake values in euthyroid research participants. All studies that determined reference values used the thyroid uptake procedure/ protocol that they were accustomed to in their respective facilities. Thyroid scintigraphy is used in hospitals for evaluation of patients with suspected thyroid abnormalities, and in some cases therapy of thyroid cancers. Commonly Technetium-99m pertechnetate and radioactive iodine imaging are used to aid in the management of patients presenting with thyroid disorders. In addition ^{123}I and ^{131}I are used for whole body imaging for follow up on patients who previously had thyroid cancers (Elgazzar, 2011).

The thyroid uptake and thyroid scan may be performed on the same day depending on which examination the patient requires. Different imaging instrumentation are used in the different procedures; however the information obtained is reciprocal. The thyroid scans are usually performed using a gamma camera, and the uptake measurement is usually obtained with a non-imaging gamma probe or a gamma camera (Ziessman et al, 2006).

Prior to thyroid uptake and scintigraphy procedures patients are routinely assessed to identify any contra indications that may affect the findings.

2.3.2 Thyroid Uptake Contra Indications

A variety of agents are known to interfere with the uptake of radiopharmaceuticals in the thyroid gland, these agents can be either in the form of antithyroid drugs or antithyroid inhibitors.
As clarified by Tripathi (2008), synthesis inhibitors are termed antithyroid drugs; however the term antithyroid drug (used to decrease the functional ability of an overactive thyroid) is applied to thyroid inhibitors. When performing thyroid uptake and scintigraphy, antithyroid drugs and thyroid inhibitors are contraindications for performing the examination as the patients would not present with a true reflection of the accumulation of radiopharmaceuticals in the thyroid.

The antithyroid drugs are classified by Tripathi, (2008) into four categories namely:

1. *Those that suppress hormone synthesis (Antithyroid drugs)*
   e.g. Propylthiouracil, Methimazole, Carbimazole.

2. *Those that suppress iodide trapping (Ionic inhibitors**)***

e.g. Thiocyanates (–SCN), Perchlorates (–ClO4), Nitrates (–NO3)

3. *Those that suppress hormone release*
   e.g. Iodine, Iodides of Na and K, Organic iodide.

4. *Those that spifflicate thyroid tissue*
   e.g. Radioactive iodine (^{131}I, ^{125}I, ^{123}I).

While establishing normal values for thyroid uptake of radioactive iodine, physicians in the reviewed literature Gonzalez et al (2002), Hooper et al (1980) and Pitman et al (1969) have taken care to exclude participants who were exposed to antithyroid drugs for the above reasons. Diagnostic doses of ^{131}I and ^{123}I have been used in previous studies to establish reference values. Other contraindications noted in the literature include history of thyroid disease, namely Hashimotos thyroiditis, toxic nodular goitre, and hypothyroidism (Culp and Huskison 1978, Bernard et al 1970, Anderson and Powsner 1996, Kauth et al 1972 and Goldberg 1964). Furthermore participants who presented with renal, cardiac and hepatic diseases were also excluded in the studies (Kauth et al 1972, Pitman et al 1969).
Although iodine is a component of the thyroid hormones it is a fast acting thyroid inhibitor (Tripathi, 2008), thus participants exposed to iodine contrast need to be excluded in thyroid uptake studies when determining normal reference values.

2.4 Serum Thyroid Hormones

There are important variables to consider when determining the reference values for thyroid uptake, more importantly a euthyroid state needs to be determined for each research participant. The indices noted in the literature to determine a euthyroid state include: TSH (serum tyrotropin), total and free T4 (thyroxine), total and free T3 (triiodthyronine), effective, thyroxin ratio (ETR), serum protein-bound iodine (PBI), free thyroxine index (T7), T3U (triiodothronine) resin uptake and level of iodine in urine and plasma Milakovic et al 2006,Hooper et al 1980 and Schober and Hunt 1976) .Not all researchers used a combination of all serum indices, different groups used a combination of tests which best suited them.
More specifically on the combination of serum tests, Milakovic et al (2006) tested TSH, total T4, free T4, total T3 and free T3. Ramos et al (2002) were the only group noted in the literature that used Technetium-99m pertechnetate to determine the reference values. This group tested the freeT4, total T4 and TSH. Hooper et al (1980) had a combination of total

T4, T3U, total T3 and FTI/T7.In addition to testing for T3 and T4, Schober and Hunt (1976) also tested the ERT, PBI including the radioactivity in plasma and urine.

In practice it is mandatory to conduct laboratory tests to confirm the diagnosis of thyroid disorders. However Sachdev (2008) emphasises that many tests used in the past are not used at the present time. Sachdev (2008) further categorizes laboratory tests into two groups; tests to determine thyroid organ functional state and tests to diagnose the cause of thyroid disease. The former was considered the best option to determine the euthyroid state in the studies to determine reference values. The most common routine tests are identified by Sachdev (2008) as TSH (to determine hypothyroid and hyperthyroid on condition that an Immuno –Radio- Metric Assay is used) total or free T4 (measured when patients are suspected of thyroid disorders) and T3 (for hyperthyroidism and disorders of iodine deficiency).

The following section on thyroid uptake measurements defines the radiopharmaceuticals, imaging protocols, equipment and formulas used to calculate the percentage thyroid uptake value.

2.5 Radiopharmaceuticals

2.5.1 Iodine -131

Historically ^{131}I was the standard radionuclide used for routine thyroid studies; however it has a disadvantage of high radiation dose to the thyroid gland caused by its long half-life (T1/2) of 8.1 days. It has beta particle emission and gamma photon energy of 364 keV however this high energy is inadequately collimated by most gamma cameras, resulting in poor quality images. The use of ^{131}I has been restricted to staging and follow-up of patients with differentiated thyroid carcinoma. Iodine-131 beta emission is successfully used for the treatment of hyperthyroidism (Christian et al, 2012).

2.5.2 Iodine - 123

Biersack and Freeman (2007) consider ^{123}I with its shorter T1/2 (13 hours) as a good substitute for ^{131}I. It is a pure gamma emitter with a principal energy of 159 keV which is suitable for conventional gamma cameras, and it does not emit beta radiation.

Kowalsky and Falen (2004) concur that ^{123}I is the imaging agent of choice and its main limitations as noted by Saha (2004) is that it is not readily available due to its expensive and complex production in a cyclotron, in addition depending on the production process chosen, contaminants such as ^{124}I and ^{125}I may be formed, increasing the dosimetry and contributing to poor image quality.

2.5.3 Technetium-99m Pertechnetate

Technetium-99m pertechnetate which is eluted from a ^{99}Molybdenum –^{99m}Technetium generator in a saline solution can also be used for thyroid scintigraphy and uptake. Technetium-99m pertechnetate is being used routinely in nuclear medicine departments internationally to assess thyroid function and uptake. Its several advantages over other iodides are listed by Christian and Waterstram-Rich (2012) as:

- Shorter half-life of only 6 hours, with less radiation absorbed dose to the patient
- Retention for a short period in the thyroid gland
- No beta particle emissions, thus allowing for a low radiation dose not just to the thyroid gland, but to the body as a whole, (10,000 times less than ^{131}I as stated by Ramos et al 2002).
- Gamma photon mono-energetic energy of 140 keV makes Technetium-99m pertechnetate ideal for imaging as it is efficiently detected by a gamma camera
- Relatively low cost and easily available
- Better image quality

Iodine - 131 was used in previous studies to determine the reference values for thyroid uptake from as early as 1959 as noted in the literature. Roche (1959) used ^{131}I to determine the reference values in isolated Venezuelan Indians. More recently Milakovic et al (2006) still used ^{131}I to determine 24 hour iodine uptake in conjunction with thyroid imaging using Technetium-99m pertechnetate. Similarly Gonzalez et al (2008) and Al-Muqbel et al (2010) used ^{131}I to determine the reference values in healthy research participants. Although researchers opted to use ^{131}I until recently to determine thyroid uptake reference values, Ramos et al (2002) was the only group that use Technetium-99m pertechnetate to determine

these values in Brazil. Their reasons for using Technetium-99m pertechnetate are in agreement with those listed above by (Christian and Waterstram-Rich, 2012).
Iodine -123 is a better substitute for ^{131}I, however only few researchers used it to determine the reference values namely Robertson et al (1975) who used a combination of ^{123}I and^{131}I, similarly Wong and Schultz et al (1977) used ^{123}I or ^{131}I, and Hooper et al(1980) opted for using ^{123}I.

The different radiopharmaceuticals used to determine the thyroid uptake values reveal the functional information of the thyroid organ. Radioiodine and Technetium-99m pertechnetate similarly provide physiological information of the thyroid organ. The following section gives an overview of the thyroid anatomy, physiology and the pharmacokinetics of radioiodine and Technetium-99m pertechnetate

2.6 Thyroid Gland

2.6.1 Anatomy

The thyroid gland is described by Hall and Guyton (2011) as a butterfly shaped organ situated immediately anterior to the larynx and trachea. Furthermore it is one of the largest glands in the endocrine system; it is highly vascular and weighs about 15 to 20 grams in an adult. The thyroid is made up of numerous follicles filled with colloid and a lining of cuboidal epithelial cells. The colloid's major element is the glycoprotein thyroglobulin containing thyroid hormones Tyroxine (T3) and Triiodothyronine (T4).

2.6.2 Physiology

Iodine is important in the physiology of thyroid hormone formation; the amount of iodine required by the body to provide sufficient amounts of hormone is 1mg/week. In this regard commonly used table salt is iodised with sodium iodide to provide sufficient iodine required by the body. Iodides taken orally are taken up from the gastrointestinal tract into the blood. One fifth of the iodides are selectively removed by the thyroid organ and used for thyroid hormone synthesis, other iodides remaining in the blood are excreted via the urine (Khanorkar, 2012).

Khanorkar (2012) further divides this formation of hormones into three phases; in the first phase the iodides are transported from the extra cellular fluid into the granular cells and

follicles of the thyroid. The iodides are then actively pumped into the cell interior (iodide pump/iodide trapping). In the second phase the iodides are converted to iodine, this process is initiated by peroxidase enzyme coupled to a hydrogen peroxide molecule. Finally the iodine couples to the thyoglobulin molecule (organification). Thyroglobulin is further released and couples with tyrosine of the thyroglobulin. Oxidized iodine in the form of a molecule will couple directly at a slow rate with amino acid tyrosine. Tyrosine is iodised firstly to monoiodotyrosine (MIT) and secondly to diiodotyrosine (DIT). Diiodotyrosine when coupled with each otherform a thyroxine (T4) molecule which stays as part of the thyroglobulin molecule. Or one single molecule of monoiodotyrosine is joined to a molecule of diiodotyrosine to form Triiodothyronine (T3).

The final stages of thyroid physiology as described by Tripathi (2008) involves the storage and release of thyroid hormones; during these two processes the thyroglobulin composed of iodinated tyrosil and thyronil residuum is transferred into the thyroid follicle cell interior and stays as thyroid colloid until it is returned into the cells through endocytosis and broken down by lysosomal proteases. The hormones T3 and T4 are finally released into the circulation while the monoiodotyrosine and Diiodotyrosineresiduum's are further deiodinated, and the iodide is further recycled.

A Summary of Iodine uptake, thyroid hormone synthesis and release is as follows:

Iodine transport, Iodine diffuses into the thyroid against a concentration gradient and is trapped.

Oxidation, Once the iodide is trapped in the thyroid cell it is converted to an oxidised form of iodine by peroxidase.

Organification, Iodine couples with tyrosine and forms MIT or DIT.

Coupling, Two DIT form T4 or one DIT and one MIT form T3.

Storage, T3 and T4 migrate to the colloid space in the middle of the thyroid follicle.

Release, T3 and T4 are secreted by reversing the storage process and reversal of the migration through the follicular cell membrane.

2.6.3 Pharmacokinetics (Radioiodine and Technetium-99m Pertechnetate)

The dissimilarity between radioiodine and Technetium-99m pertechnetate is that the former is orally administered (in liquid or capsule form) and the latter administered intravenously. Furthermore Kowalsky and Falen (2004) remark that since Technetium-99m pertechnetate has the same ionic charge and atomic radius as iodide, similarly it is taken up by the thyroid epithelial cells, in the same mechanism that they take up iodide but is not further organified. In contrast to thyroid trapping of Technetium-99m pertechnetate radioactive iodine is selectively trapped and further organified and converted into thyroid hormones, making it favourable for physiology imaging of the thyroid (Christian and Waterstram-Rich, 2012). Technetium-99m pertechnetate is taken up by the salivary glands, stomach, and a minute amount by the choroid plexus and excreted via the kidneys (Ziessmanet al, 2006).

2.7 Thyroid Imaging Protocols

2.7.1 Radioiodine Uptake Protocol

According to the SNM guidelines (2006), thyroid uptake is usually performed 18-24 hours after administration of the radioiodine. In some circumstances, it may be performed between 2 and 6 hours after radioiodine ingestion. The neck counts, background counts of the neck phantom are obtained before imaging of patients. The patient uptake is obtained by positioning the thyroid probe over the thyroid, either in supine or erect position. Uptake is usually measured with 25 - 30 centimetres between the face of the crystal and the anterior neck or phantom.

The above protocol for radioiodine uptake has been used by previous researchers to determine the reference values. Some researchers measured radioiodine uptake at 2 and 24 hours namely Harvey et al (1972) in historic literature and Gonzalez et al (2008) in more recent literature. Other groups measured radioiodine uptake at 24 hours only, Roche (1959) and Al-Muqbel and Tashtoush et al (2010), while other groups measured thyroid uptake at 6 and 24 hours; Bernard et al (1970) and Robertson et al, (1975).
The difference in imaging time is merely a difference in protocols in the different facilities. The main reason for obtaining images at 2 to 6 hours post oral administration of radioiodine reflects the iodide trapping mechanism and the organic coupling of iodine (Sachdev, 2008). Sachdev (2008) further articulates that uptake values obtained at 24 hours are progressively determined by the rate at which iodine is lost from the thyroid.

2.7.2 Technetium-99m Pertechnetate Uptake Protocol

Thyroid Uptake with Technetium-99m pertechnetate is performed 20 minutes post intravenous administration. The full syringe is measured before administration and the empty syringe is measured after injection to obtain the actual activity administered. A neck phantom is not necessary with Technetium-99m pertechnetate due to high neck and background counts obtained. This uptake procedure is performed with a gamma camera (Ziessman et al, 2006).The patient is positioned supine with the neck extended backwards and an uptake image is obtained at a distance of 20cm.

The above protocol is a standard protocol when using Technetium-99m pertechnetate for thyroid uptake calculations. This protocol was use by Ramos et al (1996) and the present study to perform thyroid uptake on research participants.

2.7.3 Instrumentation

The thyroid uptake and thyroid scan may be performed on the same day depending on what examination the patient requires. Different imaging equipment is used in different procedures however the information obtained is reciprocal. The thyroid scans are usually performed using a gamma camera, and the uptake measurement can be obtained with a non-imaging gamma probe or a gamma camera (Ziessman et al, 2006).
The most commonly used Nuclear Medicine instrumentation in thyroid uptake measurements are thyroid probe counters and gamma cameras. Routinely used instrumentation by researchers to determine the uptake values include; Thyroid probe (using ^{131}I) Kusai et al (2010), Probe counter system Nal crystal and single channel pulse height analyser(using ^{123}I) Hooper et al(1980) and a Gamma camera Low Energy High Resolution pinhole collimator using Technetium-99m pertechnetate (Ramos et al, 2002).
According to Saha (2006) the thyroid probe is the most commonly used detector to measure the thyroid uptake of ^{131}I and^{123}I after oral administration. However Khandelwal et al (2012) argue that Technetium-99m pertechnetate using a pinhole collimator is preferable as a higher radioactive dose may be administered allowing for better resolution. The disadvantage of Technetium-99m pertechnetate is the three dimensional distortion while using the pinhole collimator and the low sensitivity in the mediastinum.

2.7.4 Scintigraphic Thyroid Uptake Image

The normal anatomical scintigraphic appearance of the thyroid uptake of radiopharmaceuticals as in **Figure 2.1** (NM 0015, 2011) is a bilobal organ demonstrating homogeneous radioisotope uptake in both lobes. The isthmus which joins the two lobes medially usually demonstrates less radiotracer uptake compared to the two lobes laterally. The pyramidal lobe medially protruding from the isthmus or either of the lobes medially in some people may also be seen with less radiotracer intensity than in the two lobes (Kowalsky and Falen, 2004).

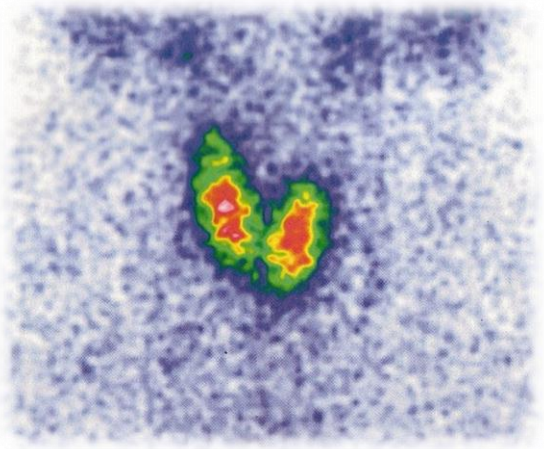

Figure 2.1Thyroid scintigraphic image

(Taken at research site, 2011)

2.7.5Thyroid Uptake Calculation and Processing

Various thyroid uptake systems, both imaging and non-imaging, are used for the measuring and registering of thyroid uptake of various radiopharmaceuticals by the thyroid gland. The different software packages usually automatically do the registration and management of the counts (thyroid, full syringe, empty syringe, background and decay time) obtained from the uptake procedure.

Theory

The calculation of the thyroid uptake index is as follows:

$$\text{Thyroid Uptake \%} = \frac{\text{Thyroid counts} - \text{Background counts} \times 100}{(\text{Pre-injection counts}) - (\text{Post -injection counts})}$$

(Mediso Clinical Processing System, 2006)

- *Thyroid* - the total thyroid counts corrected for decay
- *Background* - the total background counts corrected for decay
- *Pre-counts* - Full syringe counts corrected for decay
- *Post-counts* - Empty syringe counts corrected for decay

The decay correction is obtained by multiplying the factor (0.693T1/2) with the image counts.

- **T** is the time between the radiopharmaceutical preparation and the image acquisition.
- T1/2 is the radiotracer half-life (e.g. 6hours for Technetium-99m pertechnetate uptake)

The calculation for the thyroid uptake percentage is thus calculated using three data sets:

1. Full syringe counts
2. Empty syringe counts
3. Thyroid image counts (with a ROI drawn around the thyroid)

(Mediso Clinical Processing System, 2006)

The thyroid uptake percentage value of Technetium-99m pertechnetate may be calculated by using the following formulas and the above theory:

1. *Technetium-99m pertechnetate uptake calculation **1**:*

The Technetium-99m pertechnetate uptake can be calculated using the following relationship:

$$\text{Technetium-99m pertechnetate uptake \%} = \frac{\text{Neck Counts (cpm)} - \text{Thigh Counts (cpm)}}{\text{Administered Counts (cpm)} - \text{Background Counts (cpm)}} \times 100$$

Administered counts are obtained either by counting the tracer actually administered to the patient, with correction for decay if necessary. The thigh counts are considered as background to subtract from the neck (Thyroid) counts (SNM Guidelines, 2006).

2. *Technetium-99m pertechnetate uptake calculation* ***2***

Another formula similar to the one stated in the SNM guidelines (2006) was used by Ramos et al (1996) to calculate the Technetium-99m pertechnetate uptake percentage for each research participant and is as follows:

$$\text{Thyroid Uptake\%} = \frac{\text{Thyroid counts-Background counts} \times 100}{\text{Full syringe counts} - \text{Empty syringe counts}}$$

The dissimilarity between the former and the latter calculation are the background counts, in the first equation background counts were obtained from the thigh and Ramos et al (2006) obtained background counts from an irregular region of interest drawn just below the thyroid.

2.8 Normal Reference Value Calculation

There is contention in the literature with regards to the method of calculating the normal values for the thyroid uptake procedure. The contention is caused by the non-Gaussian data experienced by different researches. Ramos et al (1996) post plotting a frequency histogram found that the mean and standard deviation did not conform to a normal distribution curve, and the downward non-Gaussian distribution in their data was confirmed by the Shapiro-Wilksskewness test. Similarly Culp and Huskion (1978) also experienced a non-Gaussian distribution with values skewed downward. Furthermore Culp and Huskison (1978) state that using simple percentage values and two standard deviations softens the sharpness of the variance between what is normal and abnormal. Culp and Huskion (1978) conclude that the logarithmic transformation of their data produced a less skewed curve and more reliable values especially at their lower end. Likewise Harvey et al (1972) also experienced a skewed lowering in their values. The distribution of subjects in their study was not statistically normal. Therefore they concluded that the standard deviation could not be applied to determine the normal values.

The reasons noted in the literature for the skewness include; using males and females with a wide age group or using a small sample size, however Schober and Hunt (1976) argue well that many investigations with large sample sizes (600 to 1000 subjects) have also experienced the downward skewed values. Furthermore Schober and Hunt (1976) only used females in their study which also presented with a downward skewed distribution, thus they conclude that the above could not be justified as it is well known that using percentages for calculation produces skewed distributions. Schober and Hunt (1976) calculated their values using the geometric mean of the radioactive iodine uptake from the log transformation to

normalise there data, it was not possible to determine the mean and standard deviation for their study.

Nelson et al (1970) like others experienced a skewed distribution of radioiodine uptake in euthyroid subjects, and used the kurtosis skeweness test to correct their data to conform to a normal curve. The formula used was *"the square of the mean square root plus or minus two standard deviations from the mean square root";* this formula was also adopted in this study to correct for the skewed distribution.

Summary

This chapter reviewed the literature on similar studies and the results and findings of the different studies.
The literature reviewed presents similarities in research aims /objectives, namely to determine thyroid uptake normal values due to reported fluctuations in the normal thyroidal radioiodine uptake from widely separated geographic locations.
Previous studies on establishing normal values adopted a normative/norm-referenced research design. This design focuses on establishing the normal reference values for specific variables which would then serve as a guide for diagnosing and treatment planning (Portney and Watkins, 2009). Furthermore these normal values are established by taking a large group of people that fit specified criteria.

There were different approaches and sampling methods used to select the appropriate group of people that would represent a population. Three research approaches are noted in the literature; 1) Euthyroid subjects were followed up over time, 2) Medical records of patients with thyroid disorders were compared with the euthyroid group and 3) subjects were recruited from the hospital staff .The adequacy of sample size when determining thyroid uptake values was not quite clear especially when there are ethical implications with regards to sample size and age when exposing research participants to radiation. Schober and Hunt (1976) give the smallest sample size that can be used to determine reference values which gives statistically valid results as 60 subjects. The smallest sample size noted in the literature was 44 participants and the largest sample size was 671. The youngest age was 17 years and the oldest age noted in the literature was 83 years.

The thyroid uptake measurement is a common procedure performed in Nuclear Medicine. Furthermore the uptake test is useful in making a distinction between hyperthyroidism and

other forms of thyrotoxicosis or determining the dose of ^{131}I for the treatment of hyperthyroidism and the ablative dose for therapy (ARC, SNM, SPR revised guidelines, 2009). All the studies that determined the reference values used the thyroid uptake procedure or protocol that they were accustomed to in their respective facilities.

A variety of agents are known to interfere with the uptake of radiopharmaceuticals in the thyroid gland, these agents can be either in the form of antithyroid drugs or antithyroid inhibitors. All the research subjects used to determine reference values were assessed for any contraindications that may affect the thyroid uptake scan findings, for this reason an additional component was added to the normative research design by some researchers. Noted in the literature (Schober and Hunt 1976, Bernard et al 1970) was a questionnaire designed to reveal thyroid diseases and iodine contamination or ingestion.
In conjunction with medical history and clinical history examination and excluding all subjects with contraindications for thyroid uptake, serum thyroid hormone values were obtained to determine if the research subjects were euthyroid. The radiopharmaceuticals used include in more historic literature ^{131}I and ^{123}I and in more recent literature Technetium-99m pertechnetate. The different types of radiopharmaceuticals used reveal the functional information of the thyroid organ. Radioiodine and Technetium-99m pertechnetate similarly provide physiological information of the thyroid gland. The dissimilarity between using radioiodine and Technetium-99m pertechnetate is that the former is orally administered (in liquid or capsule) and the latter administered intravenously.

According to the SNM guidelines (2006) thyroid uptake of Technetium-99m pertechnetate is performed 20min post intravenous injection. The full syringe is measured before administration and the empty syringe measured after injection. A gamma camera is used with the patient positioned supine with the neck extended and an image is taken at 20 minutes following administration.
The thyroid uptake of Technetium-99m pertechnetate uptake is calculated using the following formula:

$$\text{Technetium-99m pertechnetate uptake \%} = \frac{\text{Neck Counts (cpm)} - \text{Thigh Counts (cpm)}}{\text{Administered Counts (cpm)} - \text{Background Counts (cpm)}} \times 100$$

A variety of thyroid uptake systems are used for the measuring and registering of the thyroid uptake of different radiopharmaceuticals by the thyroid gland. The general formula for calculating the thyroid uptake percentage index is as follows:

$$\text{Thyroid Uptake \%} = \frac{\text{Thyroid counts} - \text{Background counts} \times 100}{(\text{Pre-counts-post}) - (\text{Post} - \text{counts})}$$

Ramos et al (2006)

There was discrepancy in the literature with regards to the method of calculating the normal values for the thyroid uptake procedure. Different researchers used different methods to interpret the non-Gaussian data .However values are generally calculated by taking the mean of all the percentage values plus or minus one or two standard deviations (one or two standard deviations above and below the mean) to obtain the “Normal Range”.

3. CHAPTER THREE RESEARCH METHODOLOGY

This research study was conducted in order to establish normal reference values for thyroid uptake of Technetium-99m pertechnetate for the Namibian population. The research study considered a sample of participants from this population to represent the whole population Brink, (1999). The objectives of the study were; firstly to establish normal reference values of Technetium-99m pertechnetate uptake in euthyroid persons in Namibia and secondly to compare the outcome reference values to current available existing international normal reference values.

The research question enquires whether the normal reference values for thyroid uptake of Technetium-99m pertechnetate in the Namibian population differs from current traditionally used International normal reference values, furthermore the phenomenological research methodology used in this study examines the fundamental statement posed in the hypothesis that the “thyroid uptake reference values for Technetium-99m pertechnetate in a Namibian population deviate from available International normal reference values”.

There are existing International Nuclear Medicine normal reference values for thyroid uptake studies using Technetium-99m pertechnetate. The review of literature has produced recurring themes emphasising the importance of periodic check on the thyroid uptake normal values. The research strategy investigates if there is a deviation in these reference values due to geographical area.

This chapter explores the research methodology and design, its scope and limitations, the research settings, research sample and data sources, ethical considerations, instrumentation used, procedures carried out, methods of validity and reliability and method used to analyse the data.

3.1 Research Design

3.1.1 Normative Research Design

According to Portney and Watkins (2009) the intent of a normative or norm-referenced research in health related sciences is to describe standard values for the characteristics of a certain population. Furthermore this type of study focuses on establishing the normal reference values for specific variables, to serve as a guide for diagnosing and treatment planning. Portney and Watkins (2009) further explain that many tests are designed to

determine how a subject performs compared to a reference group, which is situated on an average range. Moreover these normal values are established by taking a large group of people that fit a certain criteria. As technology develops research needs to be done on the new instrumentation used to provide reference values that accurately reflects their outputs, especially in areas where different instrumentation is used to measure the same clinical values. Portney and Watkins (2009) conclude that normative studies should be large, random and represent a population's heterogeneity. Also the population of interest should be represented accurately, and replication is necessary in this type of research to clearly show the consistency and thus validate the research findings.

Previous studies(Venezuela: Roche (1959); Jamaica: Goldberg et al (1964); Alamama: Pitman et al (1969); California: Nelson et al (1970), Bernard et al (1970); San Diego: Blum and Chandra (1971), Greenspan(1970); Texas: Harvey et al (1972); Eastern Oklohoma: Kauth et al (1972); New York: Sachs et al (1972); London: Keeling and Williams (1972); Cardiff: Kirkman (1974); Vancouver: Schober and Hunt (1976); Minneapolis: Wong and Schultz (1977); Arakansas: Culp and Huskison (1978); Albuquerque: Hooper et al (1980); Boston: Anderson and Powsner (1996); Brazil: Ramos et al (1998); Sweden: Milakovic et al (2006); Santiago, Chile: Gonzalez et al (2008); Jordan: Al-Muqbel and Tashtoush et al, 2010) on establishing normal reference values all seem to have adopted a normative or norm-referenced research design. This study adopts the same approach using a norm-referenced research design. The focus of the design is to establish the normal reference values for specific variables which would then serve as a guide for diagnosis and treatment planning (Portney and Watkins, 2009). Moreover these normal values are established by taking a large group of people that fit certain criteria. To determine the thyroid uptake values Nuclear Medicine professionals were aware that a euthyroid population needs to be evaluated. In addition to adopting a normative research design scientists had different approaches and sampling methods for obtaining data from euthyroid persons.

Three research approaches are noted in the literature;

1. Prospective; euthyroid participants were followed up over time. Gonzalez et al (2008) followed up euthyroid participants over a period of twenty years while Anderson and Powsner (1996) followed up euthyroid participants over sixteen years.

2. Retrospective; a) medical records of patients with thyroid disorders were compared with the euthyroid group, Al -Muqbel and Tashtoush (2010) who as stated earlier concurrently studied patients with Hashimotos thyroiditis as well as early phase

subacute thyroiditis or b) patients records who were referred to the department for suspected thyroid disorders but were found to be euthyroid (Keelings and William, 1972, Harvey et al 1972, Greenspan 1970, Nelson et al 1970 and Robertson et al 1975).

3. Cross sectional; being the majority. Three groups; Wong and Shultz (1977) , Pitman et al (1969) and Kauth et al (1972) recruited participants from their hospital staff while Hooper et al (1980), Culp and Huskion (1978), Milakovic et al (2006), Ramos et al (2002), Goldberg et al (1964) and Roche (1959) recruited participants randomly in the community.

Schober and Hunt (1976) also opted for a cross sectional approach, however they recruited only females from the hospital staff and the community and unlike other researchers who did not state the method of recruitment in their study, they posted notices in doctors' offices inviting participants to participate in the research.

The researcher adopted a cross-sectional approach to the study. According to Babbie (2008) a cross-sectional approach to a research study allows the researcher to study a small population at one point in time and make a conclusion about the whole population. Furthermore a prospective data collection method was used. Portney and Watkins (2009) state that a prospective data collection method allows the researcher to record the research variables while the research subject passes through the evaluation.

3.1.2 Research Questionnaire

The addition of a questionnaire in this study was used to assess participants for any contra indications that may have affected the thyroid uptake scan findings. The questionnaire was specifically designed to identify research subject's ethnic groups, approximate dietary intake relating to consumption of iodine including contrast radiologic examinations and medication that would affect the thyroid uptake test. Similarly Schober and Hunt (1976) used a questionnaire to identify dietary iodine intake, medication, and any exposure to iodine contrast media. While conducting the interview using the questionnaire special attention was taken in noting different foods known to have high levels of iodine, prescribed medication and vitamins containing iodine and the use of contraceptives and hormone replacement therapy. Bernard et al (1970) also included a questionnaire in their study to obtain a true population of euthyroid persons. It was not possible in this research to use all the people who volunteered, the researcher selected the best suited volunteers by using the research

questionnaire (Appendix 1A) which aided in selecting the appropriate volunteers. Once the questionnaire was reviewed, volunteers who fulfilled the requirements were selected as research participants.

3.1.3 Non-therapeutic Research Study

The research study was a non-therapeutic research study. The MRC guidelines (2002) describe this as research with no therapeutic aspects which benefits members of the public other than the research participants. In medical research there is a wide range of studies with no therapeutic aspects. This study focuses on research using ionising radiation on human subjects. The MRC guidelines further describes 3 types of research categories when using radiation on humans namely; research on new diagnostic applications, treatment and research studies in physiology.

This study to determine thyroid uptake values can be typified as “research on new diagnostic applications”. The MRC guidelines (2002) state that the establishment of medical and biological reference values is based on an adequate selection of known normal individuals and provides standards against which abnormalities can be judged. In this study a selection of known euthyroid individuals was used to determine a normal reference range for thyroid uptake of Technetium-99m pertechnetate for the Namibian population.

3.1.4 Research dose category

When using ionising radiation on human subjects an ethical research dose category needs to be taken into consideration. In this research study 100MBq was administered to each participant. Calculations for adult radiation dosimetry appearing in The Society of Nuclear Medicine Guidelines for Thyroid Measurement (2006) gives the effective dose equivalent when using 74-370 MBq Technetium-99m pertechnetate as 0.013mSv, thus it falls under category one in the research categories (the maximum permissible dose received by a research participant not exceeding 0.5 mSv)

Due to the possibility of ionising radiation producing long-term deleterious genetic effects, the MRC guidelines Use of biohazards and radiation (2002) state that wherever practicable, research participants should be older than 40 and preferably over 50 years of age.
Medical radiation exposure is usually delivered to patients who require radiation intervention in the course of their patient management; this includes diagnostic imaging and radiation treatment. Individuals accompanying as comforters to patients are also exposed to radiation

from radiopharmaceutical administration of patients. The general public outside of these radiation facilities are also subjected to low levels of radiation if they come into close proximity with discharged patients. Other persons exposed to radiation are participants participating in biomedical research often undergoing diagnostic medical examinations involving radiation exposure that resemble the diagnostic procedures usually performed on patients (Annals of International Commission on Radiological Protection, 2007).

The MRC Guidelines (2002) on ethics for medical research (Use of Biohazards and radiation) further classifies research projects according to the categories given hereafter. The distinction between categories is related to the total effective dose received by a research subject during one year. In the first category the maximum dose permitted for a research participant should not be more than 0.5 mSv, the second category allows the annual effective dose received by a subject to be greater than 0.5 mSv but less than 5 mSv, the third category permits the annual effective dose received by a subject to be greater than 5 mSv but less than 20 mSv, and in the last category the annual effective dose received by a research participant may be more than 20 mSv but less than 500 mSv.

3.1.5 Appropriateness of Design

Historic and recent research ;Venezuela: Roche(1959)**;**Jamaica: Goldberg et al (1964);Alamama:Pitman et al (1969);California:Nelson et al(1970),Bernard et al (1970);San Diego: Blum and Chandra (1971), Greenspan(1970);Texas:Harvey et al(1972);Eastern Oklohoma:Kauth et al (1972);New York:Sachs et al (1972);London:Keeling and Williams (1972);Cardiff:Kirkman(1974);Vancouver:Schober and Hunt (1976);Minneapolis: Wong and Schultz (1977);Arakansas:Culp and Huskison(1978);Albuquerque:Hooper et al (1980);Boston:Anderson and Powsner(1996);Brazil:Ramos et al (1998);Sweden:Milakovic et al (2006);Santiago, Chile:Gonzalezet al(2008);Jordan:Al-Muqbel and Tashtoush et al, 2010) on establishing normal reference values for the thyroid uptake quantification, have all used a Normative Research approach. In all these studies, thyroid uptake was performed on euthyroid participants in selected geographical locations to determine the normal reference values.

A cross-sectional approach to the research allowed the researchers to select and study a stratified group from a population to represent the whole population. The prospective data collection method allowed the researchers to record the different variables (Age, gender, thyroid serum hormones and the percentage uptake value) relevant to the research study.

In the previously cited studies, it was still necessary for the researchers to note the clinical history of each subject to identify any thyroid disorders, iodine ingestion or contamination.

3.2 Research Setting

The research setting in this study was the Nuclear Medicine Department of the Dr Bernard May Cancer Care Centre, Central Hospital, located in the capital city of Namibia, Windhoek. The department was the only state Nuclear Medicine department receiving patients from all thirteen regions of Namibia at the time of the study. The research site was selected on the basis that it is a fully equipped Nuclear Medicine facility with the appropriate gamma camera to perform thyroid uptake scintigraphy studies. In addition the facility consists of qualified nurses trained to administer radiopharmaceuticals, a Nuclear Medicine physician, who interprets the scintigraphic studies and Nuclear Medicine technologists who perform the imaging.

3.3 Research Population

The research study had two subpopulations of interest taken from the larger population of Namibia:

1. Hospital population (including all patients referred to the state hospital's Nuclear Medicine department from all around the country).

2. Windhoek population (participants out of the public who responded to the research notices).

3.4 Research Sample and Data Sources

Due to the possibility of ionising radiation causing long-term harmful genetic effects, the MRC guidelines (2002) on the use of biohazards and radiation state that wherever feasible volunteer participants should be40 years old and preferably above 50 years of age.

When determining a sample size in research involving radiation and human subjects, the MRC guidelines (2002) advises that the number of participants participating in a study should be the minimum possible in order to obtain the required information with sufficiently small inaccuracy, particularly when participants of reproductive age are used. Neutens and Robinson (2010) define a sample as a group of people in a population that will accurately

represent the whole population. Neutens and Rubinson (2010) describe the sampling unit to be an intact group, a classroom, school, an organisation or a geographical region such as a city or country. The sampling unit selected from the sampling frame was Windhoek (Capital city of Namibia).

3.5 Sampling methods

When obtaining information about a large population special consideration needs to be taken on how the representing sample should be selected. One of the main characteristics of a sample that could have an effect on the study is the selection of the sample. Peacock and Peacock (2011) state that the sample needs to be representative of the population of interest, as it is the results from this sample selection that will be generalised to the selected research population.

In this research study there were **three sampling methods** used to recruit participants for the research study:

1. ***Purposive Sampling***

Notices in the form of flyers (Appendix 1F) were handed out randomly at the Supermarkets around Windhoek as a method of recruitment, inviting the public to participate and giving a brief explanation of the research.
The flyer invited those who were interested to send a text message to the researcher.
The message required; **VOLUNTEER NAME, CONTACT NUMBER**. The participants who responded with a text message were interviewed telephonically and selected according to the delimitations of the study.

2. ***Nonprobability Sampling***

Patients already referred to the Nuclear Medicine department with normal thyroid functions, were asked if their normal results could be used in the study (i.e., normal thyroid functions, TSH, T3, T4, and the percentage uptake of Technetium-99m pertechnetate). In this sampling method the probability that a patient would be chosen was not known, until the patient's file had been screened, and the patient's blood results were found to be normal (Neutens and Robinson, 2010).

3. **Snowball Sampling**

The identified participants possessing the requisite characteristics were given two research study notices each to give out to other persons to invite them to be included in the study

(e.g. Relatives, friends and colleagues), and these participants would in turn hand out more notices to other people in the population (Neutens and Rubinson, 2010).
Some participants put up the notices in the suburbs they live in, some sent out the notice via e-mail in the company they worked, while others used word of mouth to inform the people about the research study.

3.6 Research Sample Size

When determining sample size and age group for thyroid uptake reference values the adequacy of sample size and age group is not quite clear. Neutens and Rubinson (2010) advise that the ideal sample size should be large enough (the greater the likelihood of representativeness) to be representative of the whole population. The smallest sample size noted in the literature was 44 participants used by Robertson et al (1975) and the largest sample size noted was 671 used by Anderson and Powsner (1996).The reviewed literature has age groups ranging from minimum age of 17 by Goldberg et al (1964) who used an age group of 17 to 60 years, and a maximum age of 83 years by Robertson et al (1975) who used an age group of 21 to 83 years. In the current research described, for ethical reasons posed by the MRC, the sample size ranged from 40 to 81 years. And the sample size (81 volunteers) was determined by advice given by Brink (1999) who suggests obtaining a sample size from similar studies. Schober and Hunt (1976) suggest the smallest sample size that can be used to determine reference values while giving statistically valid results, is 60 subjects.
The difference in research design and sampling methods reviewed is not quite clear however the researcher gives possible reasons, advantages and disadvantages are as follows:

Euthyroid patients

Advantages: This group of patients although referred to the hospital for suspected thyroid disorders, are already medically proven to be euthyroid. Also these patients have already been assessed for contraindications for thyroid uptake studies, and blood values and thyroid uptake values are available.

Disadvantages: There would be several inconsistencies in the instrumentation used, the dose administered, technologist techniques, and laboratories performing thyroid function blood results.

Euthyroid participants

Advantages: Data from this population will be more accurate as care will be taken in being consistent (instrumentation, administered dose, and technologist performing the scan will be standardised)

Disadvantages: Seeking the participants from the public might not be an easy process as there are ethical implications involved, and it will be logistically time consuming and costly to select the appropriate participants according to the research criteria and performing thyroid function blood tests.

Euthyroid participants over time

Advantages: These participants provide a clear overview of the normal thyroid values over time to give an indication of how many years the values would take to significantly deviate (e.g. Anderson and Powsner (1996) who followed up euthyroid participants over sixteen years)

Disadvantages: Following up participants over time would be a difficult process as people move location. In order to determine values for a certain geographic location over time all the participants would have to remain in that specific geographic location.

3.7 Exclusion Criteria

To achieve pertinent information, certain exclusion criteria were imposed. The research study had three exclusion criteria:

1. The first criterion was for ethical considerations.

2. Second criterion further excluded participants who had factors that would affect the thyroid uptake examination.

3. The last criterion further excluded those participants that could not be included in the statistical analysis, due to abnormal scan findings.

All the volunteers who came forth had to be assessed by the first two exclusion criteria's before becoming a participant in the research study.

3.7.1 Ethics exclusion criteria

For ethical reasons the researcher had to specifically exclude those participants who were deemed to be vulnerable and needing special protection. This included persons who could not give or refuse consent for themselves, and those persons who were vulnerable to coercion or undue influence.

This included:

- Patients who cannot have a Nuclear Medicine Thyroid uptake study done, pregnant females, for the protection of the embryo/foetus (Society of Nuclear Medicine Guidelines and Annals of International Commission on Radiation Protection, 2007)
- Persons below 18 years of age
- Persons of child bearing age (18-40)
- Persons with physical or mental condition and are incapable of consenting(MRC general guidelines for Use of Biohazards & Radiation Protection, 2002)
- Prisoners (prisoners are vulnerable as they might think the research will lead to the reduction of sentence, or special privileges).
- The elderly participants who have the inability to comprehend what is entailed in volunteering as a research participant. (No research should be conducted on the elderly if the same information can be obtained from research on other adults.

Persons with mental disability include psychiatric patients or those with developmental disorders, or people who abuse substances. They may have less capacity to comprehend what the research involves and to agree to take part in it the research (MRC guidelines, general principals, 2002).

3.7.2 Thyroid Uptake Exclusion Criteria

Despite the simplicity of the Technetium-99m pertechnetate uptake test, the dynamics of the normal iodine uptake by the thyroid gland can be complicated and the factors that influence the results are widely reported Schober and Hunt (1976).To avoid the factors that could affect the thyroid uptake of Technetium-99m pertechnetate, the following participants were further excluded:

- Participants on thyroid medication (thyroid hormones or antithyroid drugs), or other medication.
- Participants who had ingested iodine rich foods 24 hours before uptake study.
- Participants recently exposed to iodinated contrast media.
- Recent administration of other radionuclides.
- Drugs affecting thyroid hormones.
- Previous ^{131}I therapy.
- Recent surgery.
- Clinical evaluation; palpable goitre or nodule.
- Incomplete clinical examination.
- Renal disease.
- Heart disease.
- Abnormal blood results

3.7.3 Statistical Analysis Exclusion Criteria

For a volunteer to be considered completely euthyroid and to be included in the population to be studied, they need to be assessed and found to be normal. A normal thyroid scan will show a normal size thyroid, in the correct anatomical position, the amount of Technetium-99m pertechnetate taken up by the thyroid will be normal according to the target to background ratio, and the distribution of radioactivity will not have areas of increased or decreased uptake. When a scan is abnormal it may either present with an area of increased uptake ('Hot' nodule), referring to a benign growth being overactive or decreased uptake ('Cold' nodule), indicating an area of the thyroid being underactive due to thyroid anomalies. All the participants that had normal thyroid hormone blood results and had had a thyroid uptake examination had to be further excluded according to findings:

- Clinically enlarged thyroid gland (one participant)
- Nodule on palpation (one participant)
- Hot or Cold nodule visualised on scan (one participant)
- Enlarged thyroid on scan (one participant)

3.8 Ethics (Use of Radiation and Human Subject Research Participants)

In general medical physicians make use of results from research in routine clinical practice. To maintain competence; physicians need to keep up with research in their respective areas of practice. Even if the physicians themselves do not engage in research, they should have the ability to interpret results from research and further apply them in practice for better patient management (World Medical Association, 2009).

Portney and Watkins (2009) assert that scientific researchers have great responsibility in all stages of the research process to protect the human rights of research participants. Health professionals who are conducting research should establish priorities and only pursue research questions that concern important health care issues. Portney and Watkins (2009) further emphasise that health professional researchers have great ethical responsibilities to conduct meaningful research; furthermore justification of a research study should be on the basis of possible scientific value of the research results.

The World Medical Association's (WMA) declaration of Helsinki (Appendix 2) states that: *"Medical research involving a disadvantaged vulnerable population or community is only justified if the research is responsive to the health needs and priorities of this population or community and if there is a reasonable likelihood that this population or community stands to benefit from the results of the research".*

Research participants make a significant contribution to medical research .Volunteers from the general public may undergo research involving radiation exposure that may be similar to the routine procedures performed on patients even though this type of research usually has no direct benefits for the volunteers. With regards to recruitment of healthy research participants the MRC guidelines general principles (2002), state that there must be no coercion, overt or covert, to induce anyone to volunteer for research. The MRC guidelines, (2002) further say that the Initial recruitment should be through circulars, notices and announcements to groups, and not by individual approach.

3.9 Informed Consent and Confidentiality

As this study required the participation of human subjects, specifically human volunteers from the general public, certain ethical issues were addressed. The consideration of the ethical issues was a requirement for the purpose of assuring the safety as well as the

privacy of the research prticipant. The important ethical issues that were taken into consideration in the research study included confidentiality and consent.
The present section in particular covers ethical issues taken into consideration in this research study, regarding research on human subjects, research on human subjects involving radiation exposure, and more specifically participants involved in radiation research that provide no direct benefit to them as research participants.

3.10 Research Authorization

Before the research was conducted the proposal for the research was submitted to the Cape Peninsula University of Technology's Faculty of Health and Wellness Sciences Ethics committee to ensure non infringement of human rights. This research study also had to obtain permission from the host country (Namibia) and institute (Ministry of Health and social services, Dr B May Cancer Care Centre, Nuclear Medicine Department and the Namibian Institute of Pathology)

The following letters were received in response to ethics application for this research study:

- Ethics Approval Certificate, Cape Peninsula University of Technology's Faculty of Health and Wellness Sciences (Appendix 3A).
- Approval letter from Ministry of Health and Social Services research committee (Appendix 3B).
- Approval Letter from Namibia Institute of Pathology (Appendix 3C).
- Approval letter from Radiation Protection (Appendix 3D).

3.11 Research Ethics Declaration

The World Medical Association-WMA, (2009) has formulated the Helsinki Declaration (Appendix 2) as a statement of ethical principles for research involving human participants, including research on human material and information. In this research study involving human subjects the principal researcher adopted the principals and adhered to the guidelines accordingly (WMA Declaration of Helsinki, 2009).

Thus taking into consideration the above mentioned principals and guidelines there was a fully descriptive consent form (Appendix 1B) explaining in detail what the research entailed.

The consent form was translated into Afrikaans for those participants that were not fluent in English, and signed by the research participant, researcher and the translator. Furthermore all appendices with participant data had a numbering code and not the participant's names. All information obtained on the consent form was treated confidentially.

The WMA emphasises that when conducting research on human subjects, research should be conducted by professionals with the appropriate training and qualifications. Also the participants require supervision by a competent qualified physician or health care professional, as the responsibility for the protection of research participants, although they have given consent always rests with the qualified professionals. The following qualified professionals assisted in the research study; a registered nurse trained to inject radioactivity, administered the radioactivity, a qualified Nuclear Medicine technologist (principal investigator) performed the thyroid uptake study and a qualified Nuclear Medicine physician processed the thyroid uptake study, determined the uptake value for each participant, and was on standby for any emergencies.

Information given to Participants (Blood & Radioactivity)

In order to secure the consent of the selected volunteers, the researcher provided all the important details of the study, including the study's aim and objectives. By explaining this important information, the volunteers were able to understand the importance of their role in taking part in the research study. The volunteers were informed about the Thyroid hormone (TSH, T3 and T4) values that would be obtained from the blood samples, the amount of blood to be drawn (4ml), and the location of blood withdrawal (Left/Right antecubital fossa or Left /Right hand). With regards to radioactivity the volunteers were informed about the type of radioactivity (Technetium-99m pertechnetate) to be injected, the dose (100MBq), possible side effects, and the imaging procedure with the gamma camera. Furthermore the volunteers were informed about radiation exposure and radiation protection. To ensure that the selected research participants were not exposed to other unnecessary radiation handled in the department on the daily basis, no other radiation sources were handled in the department except that to be injected.

To protect the human rights of the participants the researcher strictly adhered to the MRC guidelines (2002) on medical research regarding the use of radioactive nuclides which include:

- Only qualified personnel assisted in this research study as irradiation of research subjects should be undertaken only by qualified and trained professionals.

- Ethics certificate was granted from the Research Ethics Committee. prior to carrying out the research study.

- Approval should be based on the advice of an appropriate expert committee, and be subject to national and local regulations, thus all expert committees provided their advice, and the local and national regulations were adhered.

- A consent form was available for each research volunteer allowing each participant to fully participate by free will and sign informed consent forms.

- The radiation risk was explained to all participants on the consent form and the participants were informed that the radiation risk was 0.013mSv and is within the acceptable research dose limits. (The recommended annual dose limit for general public may be greater than 5mSV but less that 20mSV (ICRP (2007) – International Commission on Radiological Protection).

- The magnitude of risk to research participants was evaluated by all authorities involved in the research study and authorisation to conduct the study was obtained

- A calculation of the absorbed dose was determined with the assistance of professionals in the field, and the results documented. the administered dose used in the present study was 100MBq. Calculations for adult radiation dosimetry appearing in The Society of Nuclear Medicine Guidelines for Thyroid Measurement, (2006) gives the effective dose equivalent when using 74-370 MBq Technetium-99 m pertechnetate as 0.013mSv.

3.12 Data Collection Methods and Instruments

Participant Demographics

The research participants biographical information collected in this research study was:

- Age
- Gender (male/female)
- TSH blood levels
- Free T3 blood levels
- Free T4 blood levels

- Percentage thyroid uptake of Technetium-99m pertechnetate

Age and gender

The age and the gender of each participant were obtained by verbal communication.

Blood sample (TSH, T3, and T4)

Once a volunteer had been evaluated with the questionnaire designed to reveal thyroid diseases and iodine contamination or ingestion, and was identified as a suitable volunteer, a blood sample was obtained from each volunteer and sent to the laboratory to determine if the thyroid hormone functions were normal.

Participants Health

Each participant had their own individual sterile research pack (**Figure 3.1**) containing the following items:

- Eluvial
- 2ml syringes
- 0.5 blue needles
- 5ml syringes
- 0.5 green needles
- 5 cotton pads
- Plasters
- Gloves
- heparin tubes

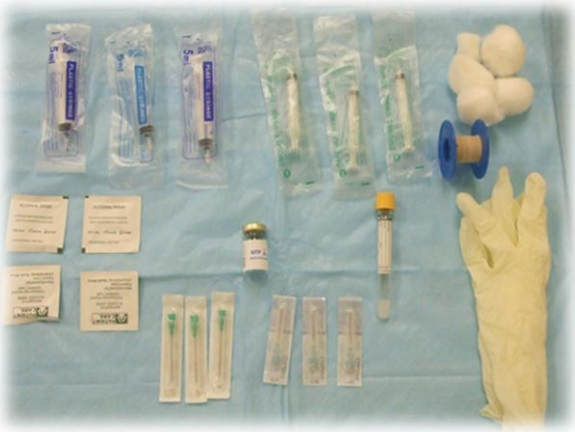

Figure 3.1 Participants research pack

(Picture taken at research site, 2011)

Collection of blood

The blood was collected using a sterile heptane tube and green needle (**Figure 3.2**)

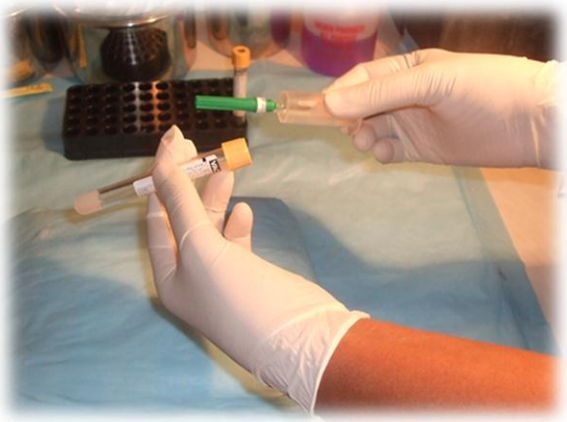

Figure 3.2 Heparin tube with 0.5 green needles

(Picture taken at research site, 2011)

Blood was drawn intravenously from Left/right antecubital fossa (**Figure 3.3**) or Left/Right hand (Figure 3.4). The blood was drawn once only.

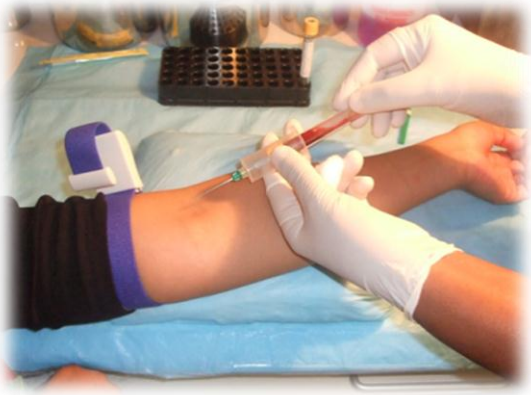

Figure 3.3 Blood drawn from Left/Right antecubital fossa

(Picture taken at research site, 2011)

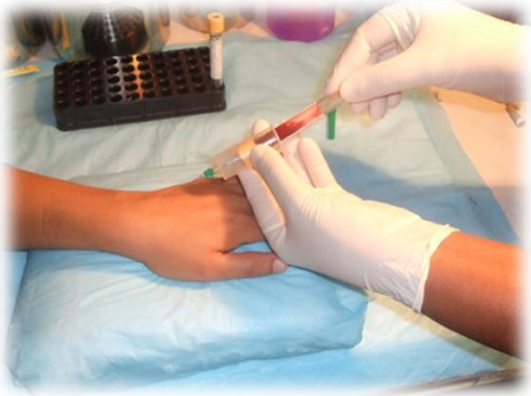

Figure 3.4 Blood drawn from left /right hand

(Picture taken at research site, 2011)

The blood sample size was 4ml in a heparin tube (**Figure 3.5)** labelled with participant code (e.g.NM001) and sent to the laboratory for further processing to obtain thyroid function values of the TSH, T3 &T4 for each participant.

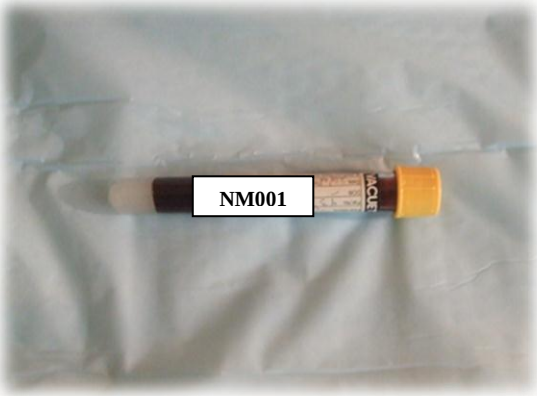

Figure 3.5 Heparin tube labelled with volunteer code

(Picture taken at research site, 2011)

Percentage thyroid uptake

The radionuclide used in this study was Technetium-99m pertechnetate. Technetium-99m pertechnetate was eluted from a ^{99}Mo- ^{99}Tc generator, in a saline solution and was used in this chemical form i.e. Technetium-99m pertechnetate.Technetium-99m has mono-energetic gamma photon energy of 140 keV which makes it ideal for gamma camera imaging .It has no beta particle emissions. Upon isomeric transition Technetium-99m is not converted into another element, it simply decays to Technetium-99 which is the ground state of the same radionuclide and remains Technetium (Saha, 2004).
After intravenous administration the thyroid follicular cells will trap the Technetium-99m pertechnetate in the same way it traps iodide, and is not metabolised further than the thyroid trapping mechanism (Kowalsky and Falen, 2004).

Method of radiopharmaceutical administration

A 2 ml syringe (**Figure 3.6**) with a 0.5 green needle (**Figure 3.7**) was used to administer the radioactive dose.

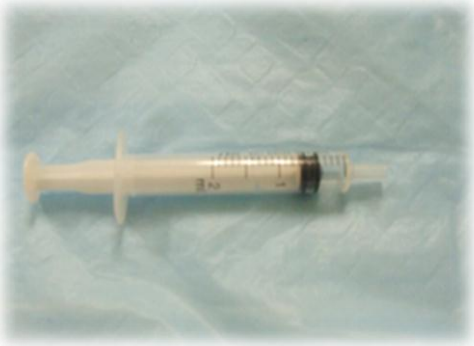

Figure 3.6 2ml syringe used for dose administration

(Picture taken at research site, 2011)

Figure 3.7 Green needle used for dose administration

(Picture taken at research site, 2011)

The radionuclide was administered intravenously into the Left/Right antecubital fossa (**Figure 3.8**) or Left/ Right hand (**Figure 3.9**), Frequency= injected once only)

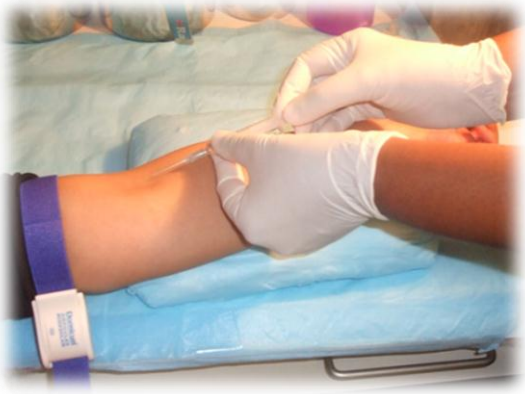

Figure 3.8 Radionuclide administration in left antecubital fossa

(Picture taken at research site, 2011)

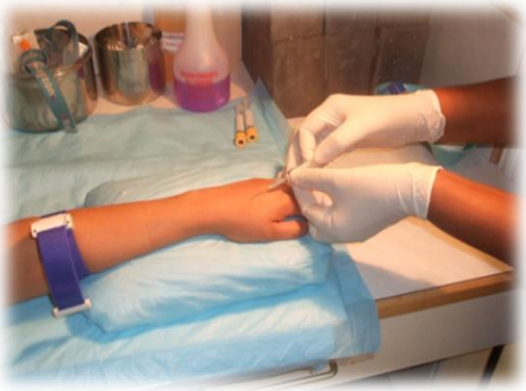

Figure 3.9 Radionuclide administration in left hand

(Picture taken at research site, 2011)

The dose of radioactivity administered to each participant was 100MBq (74- 370MBq) as stated in the ACR-SNM-SPR (2009) procedure guidelines for thyroid uptake measurements. The volume of radiopharmaceutical depended on the concentration of the radioactivity in the syringe but was generally less than 2ml **.**The dose administered was measured before and after it has been injected in order to obtain the counts for the exact amount administered. Furthermore the injection site was imaged to determine any residual activity at the site of injection to more accurately determine the percentage uptake in the thyroid gland.

Imaging procedure

Once a participant had been injected and the empty syringe measured, he/she would wait for 20min duration for the radionuclide to localise in the thyroid. After the 20min waiting

period the participant was placed under the gamma camera (**Figure 3.10**) in a supine position, with a pillow under the shoulders to extend the neck. The detector head of the gamma camera was positioned as close as possible over the anterior neck (thyroid centred) and the thyroid uptake view was obtained using the departmental thyroid uptake imaging protocol and a Low Energy High Resolution collimator.

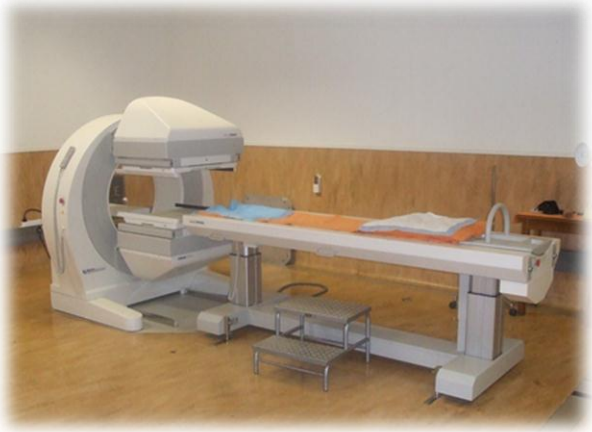

Figure 3.10 Gamma camera used to image participants

(Picture taken at research site, 2011)

The following views/projections were obtained for each individual.

- Full syringe with radioactivity dose before administration
- Empty syringe after administration of radioactivity
- Injection site (to determine any misadministration)
- Anterior thyroid view 20min after radioactivity administration

Calculation and processing method of Technetium-99m pertechnetate:

The methods used for the calculation of thyroid uptake of Technetium-99m pertechnetate uptake calculation are based on thyroid gland image, full syringe counts before radiopharmaceutical injection and empty syringe counts post injection. The total number of counts was determined by an irregular region of interest drawn around the boarders of the thyroid gland tracking the thyroid gland as close as possible (TH) see **Figure 3.11.** Another rectangular region of interest was drawn under the thyroid gland region of interest with a width approximately equal to the gland and representing the background (BK). The full syringe counts (F) before injection and empty syringe counts (E) after injection are obtained from the images.

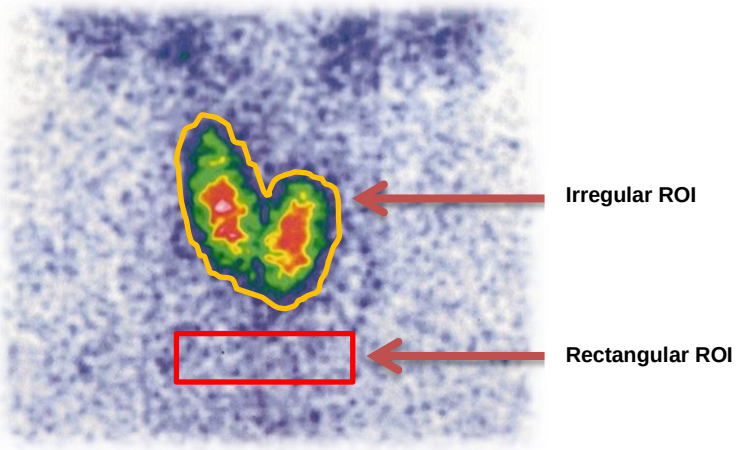

Figure 3.11 Irregular ROI around the thyroid gland and rectangular background ROI und the thyroid gland

(Picture taken at research site, 2011)

All the counts obtained were corrected for the decay of Technetium-99m pertechnetate and acquisition time. The thyroid uptake of Technetium-99m pertechnetate for each volunteer was calculated using the following formula:

$$\text{Thyroid Uptake \%} = \frac{\text{TH (thyroid counts) -BK (background counts)} \times 100}{\text{F (Full syringe counts) - (Empty syringe counts)}}$$

The average time taken to process two volunteers was 60 minutes, taking into consideration full explanation of the research procedures and completion of all the necessary documentation.

Data Capturing and Handling

To further produce research results that are valid and reliable for statistical analysis, correct capturing and handling of data was done and checked for errors in the data collected (Peacock and Peacock, 2011).
There was several data capture sheets which were designed to aid in reaching the research objectives. Each vparticipant had a research file with a unique code for data capturing purposes.

The participant research file consisted of:

1. Data capture sheet (Appendix 1C)

2. Research methodology checklist (Appendix 1D)

3. Questionnaire (Appendix 1A)

4. NIP Blood form (Appendix 1E)

The participant files were all in paper format and were stored initially at the research site and later transported to Cape Town for data analysis.
The advice of Peacock and Peacock (2011) was followed and the paper and data forms were handled as follows:

1. The folders were filed systemically (NM001 to NM083) to allow easy retrieval at a later stage

2. The data was stored in the department safe, complying with the data protection requirements. Only the researcher had access to the safe.

3. The personal details (Name and contact number) were recorded in a separate logbook and stored separately from the data forms for confidential purposes.

4. A copy of all the patient files was obtained and stored as the data was transported to Cape Town, and possible loss of data could occur.

5. The data will be stored for five years.

3.13 Research Validity and Reliability

There are two aspects that need to be taken into account when working with measurements in clinical research. At the core of measurement in clinical research is the first prerequisite; reliability, which is described by Portney and Watkins (2009) as the extent to which the measurement is consistent and does not produce error. The second prerequisite is validity, which makes sure that the instrument is measuring what is expected to be measured.

According to Jackson (2008) there are two types of measurement errors. These measurement errors are grouped into trait errors and method errors. Trait errors are caused by the research participants, and the method errors arise from the researcher, practitioner, or the research environment.
Error arising from research participants/ research participants tend to be more difficult to control, while errors caused by the researcher, practitioner or environment can be reduced, by providing training on how to use the measuring instrument and how to perform quality

control on the instrument. To minimise trait and method errors the researcher conducted a pilot study, selected appropriate qualified research assistants and performed quality control on the research instruments.

Pilot study

A pilot study was carried out at the research site to test the questionnaire. The survey questionnaire was used as the main tool to select the participants that would provide data that was pertinent to the study. The questionnaire was divided into two main sections: a section for medical history and a section for dietary iodine. The medical history section explored any medical contraindications for the thyroid uptake procedure and the dietary iodine section explored the intake of different types of foods containing iodine to get an indication of dietary iodine intake. In order to test the validity of the questionnaire used for the study, the questionnaire was tested on 15 patients. These patients as well as their answers were not part of the actual study process and were only used for testing purposes but were used to modify the questionnaire. The questionnaire was revised after identifying important questions that were overlooked, and excluded questions that did not aid in the research survey.

For ethical reasons the pilot study did not extend to further image the patients according to the research pre-set protocol, However the research assistants recorded patient data (while patient passed through the department) on the research data sheets to familiarise themselves with the data capture process.

Measuring Instruments

The following measuring instruments were used in this research study:

- The Gamma Camera (used to calculate the percentage (%) uptake of Technetium-99m pertechnetate) used in this study underwent calibration and daily uniformity tests to ensure that it would measure the thyroid uptake of Technetium-99m pertechnetate accurately.

  A pre-set research protocol titled **NUCLEAR MEDICINE RESEARCH** was set up on the camera to ensure that there was consistency with each participant being imaged. The pre-set protocol had a code (**NMR**) that was put into the computer prior to imaging of each participant. Only one gamma camera was used on all participants.

- A Dose Calibrator (used to assay the dose for each volunteer) was calibrated to make sure that its dose measurements were accurate. Inspection was carried out to make sure that the appropriate quality control procedures were performed on the dose calibrator to ensure research validity and reproducibility.

- Time Clock (used to record the time the dose was drawn up, the time of empty syringe measurement, and time of injection and the time of uptake imaging). The same digital clock was used in this research for consistency purposes to record the different times.

To further ensure research validity and reliability selected qualified staff members of the Nuclear Medicine department were involved in the research study as research assistants. Although all staff members are acquainted with the thyroid uptake examination done in the department further individual induction and training was necessary. The allocation of tasks was permanent throughout the research to ensure consistency; furthermore the principal researcher viewed and checked the data capturing sheets, for each participant.

3.14 Data Analysis

The statistical program used in this research for data entry (using research codes for each prticipant, **NM 001** to **NM 076**) and data capture was an excel spread sheet, The thyroid uptake values of Technetium-99m pertechnetate were calculated for the 76 participants. The uptake values were further divided into different groups; male, female, ethnic groups and euthyroid group (including all males and females).
The excel software program was used to calculate the mean and the standard deviations for the different groups. The statistical limits of normalcy were determined by two methods namely; by obtaining the mean plus or minus two standard deviations (i.e. 95.5% confidence interval) and using the 5^{th} and 95^{th} percentile.
Analysis was separately conducted for the euthyroid group (male and female participants) using the international reference values. The former and the latter were compared using the one way analysis of variance (ANOVA).The limits of normalcy were displayed in an error bar graph, The error bar was selected as it presents the mean and the error bar indicating one standard deviation above and below the mean.

3.15 Role of the Researcher

The researcher took into consideration the objectives of the research study and selected the appropriate method of data collection and analysis to be adopted. Furthermore the researcher established a sample design that would be the primary source of information and selected the tools for the data collection.

The researcher was actively involved in the research process by evaluating each participant according to the research delimitations and selecting the appropriate euthyroid participants. The researcher recorded the necessary data (age, gender, thyroid serum hormone values, and percentage thyroid uptake) as the participants went through the evaluation.

To prevent selection bias, as the researcher had to select the appropriate participants for the study, the researcher provided a clear definition of the study population. Information bias was prevented by using a standard measuring instrument (gamma camera) and testing the questionnaire on a small pilot group.

3.14 Research Time Line

Table3.1 Research Time Line

	ACTIVITY	TIME (months/weeks)	Duration
1	Proposal writing	3 months	February 2011 – April 2011
2	Obtained ethical approvals	2 months	June 2011 – July 2011
3	Pilot study	2 weeks	1 August-17 August 2011
4	Data collection	4 months	August 2011-December 2011
5	Data analysis	3 month	January 2012 - March 2012
6	Writing up	6 months	March 2012 - August 2012

Summary

This chapter has provided the reasons for selecting the chosen research methodology and how the researcher implemented this methodology. A Normative Research Design was chosen to conduct this research as it was determined to be a useful design for establishing the normal reference values of thyroid uptake of Technetium-99m pertechnetate for the Namibian population. To establish the normal values it was important to use a research method that would allow the researcher to obtain the data pertinent to the research and to calculate and determine the values. The data was collected from two subpopulations of interest taken from the larger population of Namibia; the Hospital population (including all

patients referred to the state hospitals Nuclear Medicine department from all around the country) and the Windhoek population (volunteers out of the public who responded to the research notices).
Three sampling methods (Snowball, Purposive and Non-probability) were used to obtain euthyroid participants to represent the population. The thyroid uptake was obtained from the euthyroid participants from the two sub population groups; the hospital population, including all patients referrals to the department and volunteers from the general public. The normal values for the 76 euthyroid participants were determined using two methods namely; by calculating the mean plus and minus two standard deviations and using the 5^{th} and 95^{th} percentiles.

The following chapter will present the findings of the data obtained from the research participants used in this research study

CHAPTER FOUR RESULTS

Eighty three participants considered to be euthyroid from Windhoek general population including two euthyroid thyroid patients selected from the patient population from Windhoek State hospital received Technetium-99 m pertechnetate as part of their thyroid evaluation.

The results of seven subjects were excluded due to the following reasons;

- NM030 – Abnormal blood results *(participant had hypothyroidism),* TSH>100, T3 2.88, T4 0.06
- NM036 – Abnormal blood results (TSH -2.88, T3 4.28, T4 10.48)
- NM 023 – Misinjection
- NM078 – Blood lost in lab
- NM065 – Empty syringe not recorded
- NM083 – Participant did not complete study
- NM058 – Participants scan showed an enlarged thyroid with a “hot” nodule

The total study sample size comprised of 76 research participants (58 females and 18 Males).Because of Radiation safety concerns the age range was 39 years and above. The participants were selected from a population of mixed ethnicity consisting mainly of Hereros, Ovahimbas, Kavangos, Caprivians, Ovambos, Damaras, Namas, Bushmen, Coloureds, Tswana and Caucasians. Ethnic distribution of the 76 participants are illustrated in **Figure 4.1** include 40.8% (n=31) Ovambos, 23.7 % (n=18) Hereros, 14.5% (n=11) Coloureds), 7.9%(n= 6) Namas, 6.6% (n=5) Damaras and others (4 Caucasian’s and 1 Foreigner) 6.6%

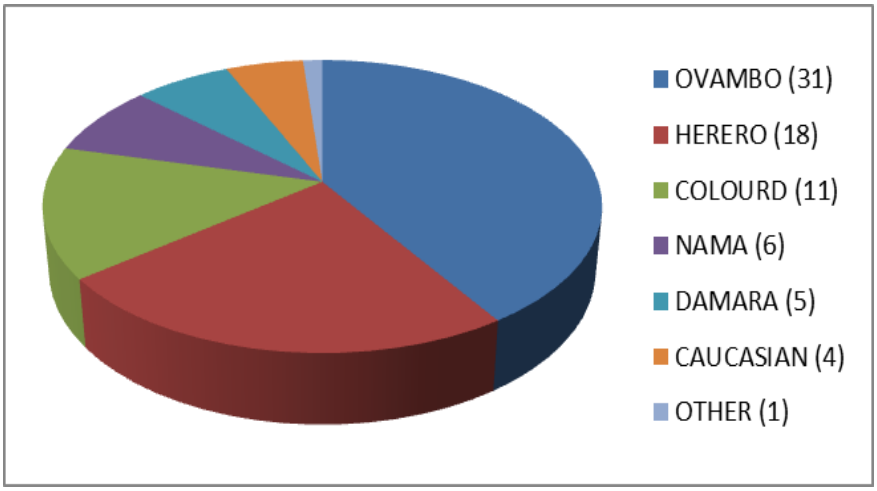

Figure 4.1 Ethnic group distribution of study sample

During clinical evaluation of the 76 euthyroid participants used for statistical analysis, no enlarged thyroids or nodules were noted. These findings were further confirmed by the scintigraphic image findings which showed no thyroid enlargements or nodules for all 76 participants. Furthermore, when Technetium-99m pertechnetate uptake values for the participants were evaluated against the values used in the department (0.75% to 4%) there were no uptake values above 4%. However 51% (n=39) participants had an uptake percentage of Technetium-99m pertechnetate lower than 0.75% and the remaining 49% (n=37) had uptake percentages within the departmental values.

The interview questionnaire used in the study had two sections namely medical history and source of dietary iodine. Analysis of the medical history noted that participants had not had any thyroid disorders in the past or suspected of having any clinical evidence of current thyroid disorders. No participant had been exposed to iodinated contrast media or radionuclides during the time of the evaluation or in the previous year before the investigation. There was no history of heart medication having been taken within the last two years by any participant. None of the female participants had used any iodine containing vaginal douche and none of the female participants were pregnant or nursing at the time of investigation. None of the participants were smokers.

The different ethnic groups all have unique diets .Analysis of the source of dietary iodine presented found that all participants used iodized table salt on a daily basis while cooking; sea food was part of the diet as all participants ate fish at least once a month. Twenty three participants took multivitamins and minerals on a daily basis and these include; Q 10 Fatigue,Dynamisan, Pharmaton, Glucosamin, Sentrum, Bico, Vitamin C, Beta Vita, Vitathion,

Vitamin A to Z, and Vital. Twenty of the participants were taking energy supplements on a daily basis. All participants consumed fresh milk in their tea or coffee on a daily basis. Cheese, yogurt, ice cream, frozen yogurt and eggs were only taken occasionally as a luxury and all participants consumed at least two slices of bread a day. From the analysis of dietary iodine section of the questionnaire we can conclude that the main source of dietary iodine for this population is iodized table salt, dairy products and bread.

The serum thyroid hormone values had to be determined for each participant. The results of the serum thyroid hormone values are summarised in **Table 4.1**.All the thyroid hormone values were within the normal range of the laboratory used in this study.

Table 4.1 Thyroid functions in normal participants

Group	Total	TSH Mean±SD	T3 Mean±SD	T4 Mean±SD
Females	58	1.58±0.76	4.82±0.80	10.36±1.84
Males	18	2.06±1.40	4.96±0.79	10.17±1.77
Euthyroid	76	1.70 ±0.96	4.86±0.79	10.32 ±1.82
Normal Range		0.34- 5.60µIU/mL	3.80 - 6.00 pmol/L	7.2 – 21.1 pmol/L

The actual sample size for the research study was 76 euthyroid participants. The characteristics and % uptake results of the 76 euthyroid participants at the Windhoek Central Hospital's Nuclear Medicine Department are summarised in **Table 4.2**. The mean Technetium-99m pertechnetate uptake for the euthyroid group, male group and female group were 0.78 ± 0.45, 0.65± 0.38 and 0.81± 0.47. All three groups male, female and euthyroid group had a 95.5% confidence interval (CI).

Table 4.2 Means and Ranges of Technetium-99m pertechnetate uptake in males females and euthyroid Namibian participants

Group	Total	Percentage (%)	Mean Age (y)	Age Range (y)	% Uptake range	% Uptake Mean ± SD	% Uptake 95.5 % CI
Males	18	23.7	49.83	39 - 67	0.31 – 1.07	0.65 ± 0.38	0.50 - 0.88
Females	58	76.3	49.28	41- 81	0.34 – 1.28	0.81 ± 0.47	0.68 - 0.93
Euthyroid	76	100	49.41	39-67	0.31 – 1.28	0.78 ± 0.45	0.33 – 1.23

The gender comparison of the normal values found in this study are compared to the whole euthyroid group (males and females) and illustrated in **Figure 4.2** with normal values lower in the males than the females.

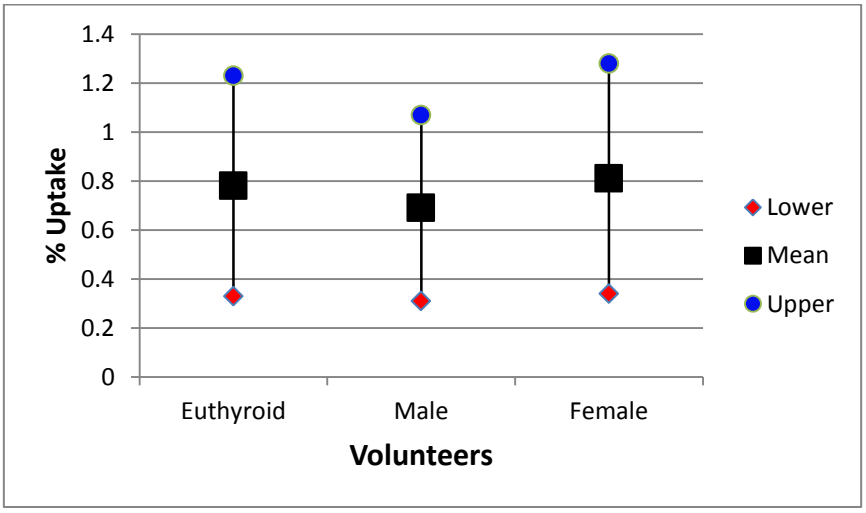

Figure 4.2 Gender comparison of normal values found in this study

The 76 euthyroid participants were divided into the ethnic groups. Although the sample sizes for the different ethnic groups were small the normal values were calculated and illustrated in **Figure 4.3.**

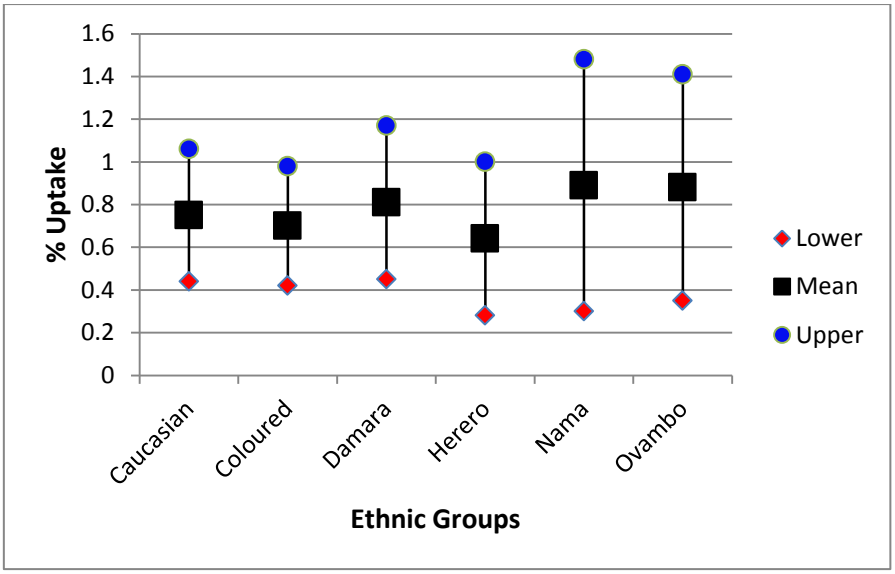

Figure 4.3 Ethnic comparison of normal values found in this study

A scatter graph of the Technetium-99m pertechnetate uptake values for all 76 participants is illustrated in **Figure 4.4.** Twenty six percent of the participants (n=20) had extremely low

Technetium-99 m pertechnetate uptake (< 0.5). The cause for these extremely low uptake values is unknown.

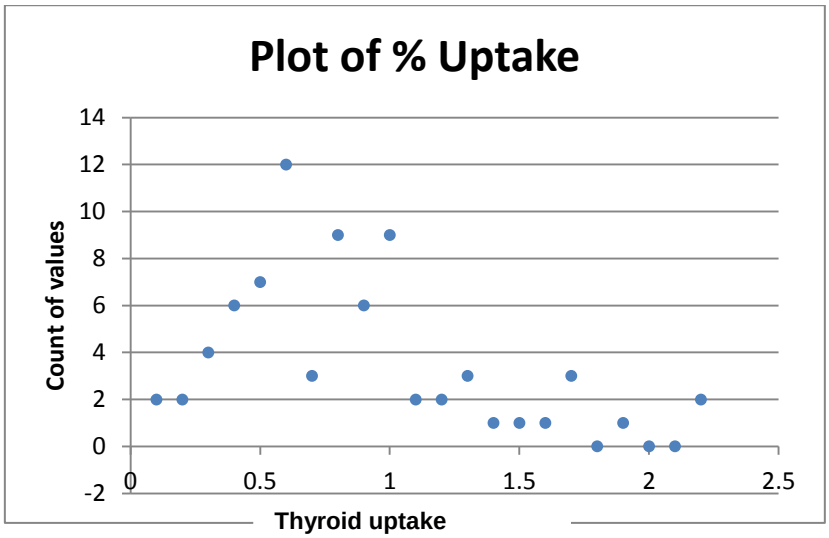

Figure 4.4 Technetium-99 m pertechnetate uptake values for all 76 participants

When Technetium-99m pertechnetate uptake values for the participants were compared to the values used in the department (0.75% - 4%) there were no uptake values above 4%. However 52% (n=39) participants had an uptake percentage of Technetium-99m pertechnetate lower than 0.75% and the remaining 49% (n=37) had uptake percentages within the reference values used.

The Kolmogorov-Smirnov[a]Lilliefors Significance parametric test presented us with results that state that our population has a non-normal distribution, and therefore the standard deviation should not be applied.

When the uptake values were plotted on a normal distribution plot, the uptake values for the 76 participants presented a skewed distribution (illustrated in **Figure 4.5).** For a population to be normally distributed the data obtained should display a straight line. When data obtained deviates from the straight line, it denotes that the particular population being researched is abnormally distributed. The skewed distribution illustrated here has also been reported in older studies (Bernard et al, 1970, Harvey et al 1972, Pitman et al 1967, Nelson et al, 1969).

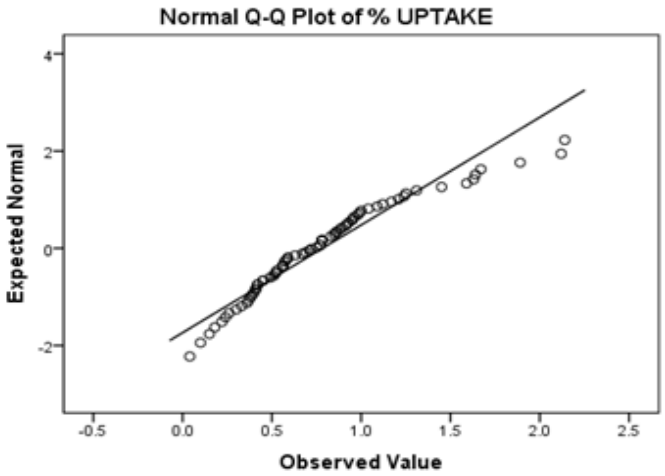

Figure 4.5 Uptake Values of 76 participants plotted on a normal distribution plot

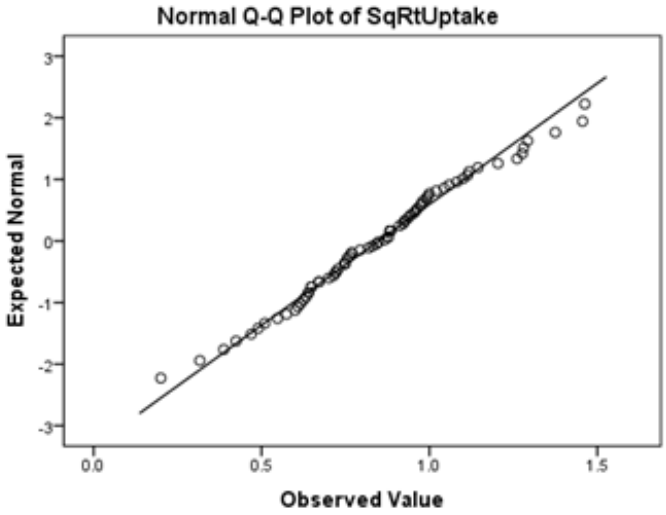

Figure 4.6Skewness corrected on the normal distribution plot

The *statistical test for skewness and kurtosis* formula used by Nelson [6] was adopted in this study and **Figure 4. 6** illustrates the skewness corrected. When the data was corrected for skewness the normal values were 0.35% to 1.22% (95.5% CI) of Technetium-99m pertechnetate at 20 minutes.

This skewness is further demonstrated in the frequency histogram in **Figure 4.7** which presents the frequency distribution of Technetium-99m pertechnetate uptake at 20 minutes in the 76 euthyroid participants compared with the normal range routinely used in the department (0.75% - 4%). The shaded bars illustrate participants with uptake values falling below the department's lower limit of normal (0.75%).As shown 52% (n=39) of the

participants had less than 0.75% uptake at 20 minutes, these uptake values would have been noted as abnormal according to the previous departmental normal values.

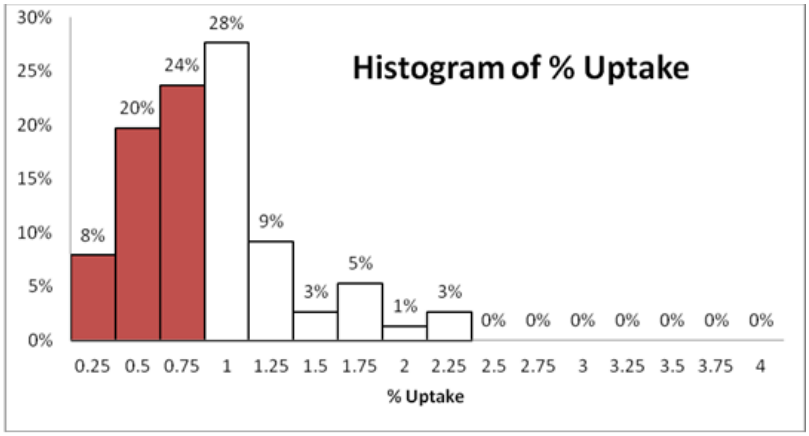

Figure 4.7 Relative Frequency Histogram of percentage uptake showing the old Namibian values.

An alternative method to determine the normal reference range for Technetium-99m pertechnetate in a study sample obtained from a non- normal distribution was determined using the 5th and the 95th percentiles (**Figure 4.8).** The normal reference range using the 5th and the 95th percentile was 0.17% - 1.7%.

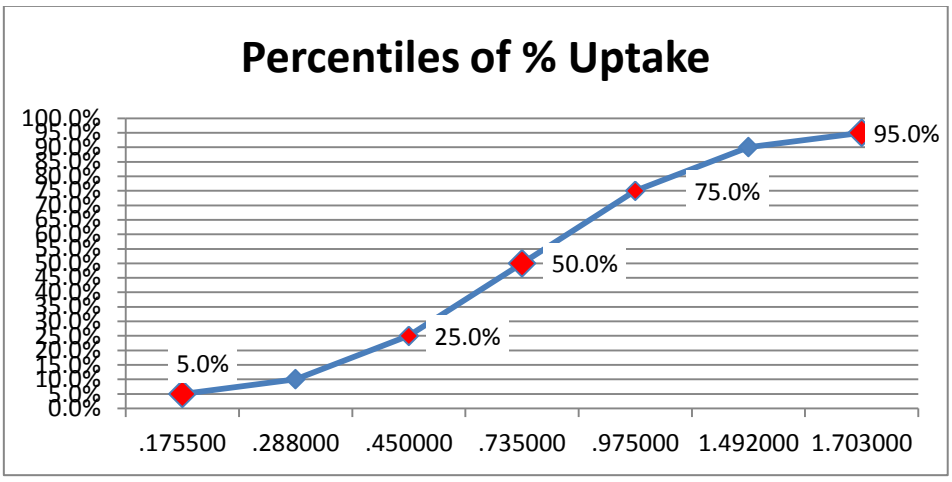

Figure 4.8 Cumulative percentage polygon presenting the percentiles of the percentage uptake values

CHAPTER FIVE DISCUSSION

Thyroid scintigraphy and thyroid uptake quantification is performed routinely in Namibia. There are several definite indications for the thyroid uptake procedure including but not limited to making a distinction between hyperthyroidism and other types of thyrotoxicosis and determining the dose of ^{131}I for the treatment of hyperthyroidism or the ablative dose for therapy (ARC, SPR, SNM revised guidelines, 2009).

In Namibia the reference range used for thyroid uptake of Technetium-99m pertechnetate is 0.75% to 4 % of the administered dose at 20 minutes. These ranges have been adopted from international Nuclear Medicine guidelines on thyroid uptake measurement, as it is not always convenient for each Nuclear Medicine facility to determine Technetium-99m pertechnetate uptake in normal euthyroid individuals. There has been no specific reference range established for this geographical region, despite the fact that all the changes from historical data to more recent data proves that there are geographic variations as well as changes in iodine intake between populations. It is thus necessary to revise these normal reference values for correct interpretation of the thyroid uptake procedure.
To correctly interpret the thyroid uptake examination one needs to know the range of values to be anticipated in normal persons with ordinary diet habits and no history of iodine ingestion from medication or contrast media. Nevertheless the iodine intake may fluctuate without public consciousness, thus the greatest pitfall in the thyroid uptake test is the omnipresent iodine atom present in our daily lives. For this reason the reference ranges for thyroid uptake of Technetium-99m pertechnetate for the Namibian population were reviewed in Windhoek between September 2011 and November 2011.

Ethnicity and Geographic Variation

The Nuclear Medicine Department at the Windhoek Central Hospital was the only state Nuclear Medicine department at the time; patients referred to this Nuclear Medicine department come from all thirteen regions of Namibia. The regions are comprised of rural, urban, dry savannahs, and mostly dessert. Namibia consists of mixed ethnic groups constituting 50% Ovambo , 9% Kavangos, 7% Hereros, 7% Damaras,5% Namas,4% Caprivian, 3% Bushmen,2% Coloureds , 0.5% Tswana making up 87.5% while others (Caucasians, Ovahimba, foreigners that have settled in Namibia) constitute 12.5% of the population All ethnic groups have their different unique diets. Although the majority of

population usually migrate to this region for employment, the Ovahimba, Kavangos, Caprivians, Bushmen and Tswana ethnic groups were not represented amongst the research subjects.

Iodine in Namibian Diet

Despite the wide variations of the thirteen regions, the Namibian population has sufficient dietary iodine intake. Similar to all other developing countries the population used iodized table salt and other forms of iodine rich foods noted in the diet included dairy products, sea food and bread. Besides table salt, sea food and bread, other sources of iodine not noted in the present study make it difficult for nutritionists to anticipate the level of iodine consumption in the future. It is well known that iodine content in foods varies with geographic location, thus the iodine content of food from one country or geographical location cannot be universally used for estimating the iodine consumption of people living in different geographical locations (World Health Organisation, 2007).The uptake of radioactive iodine tends to be lower in geographic areas with sufficient amounts of dietary iodine than in areas where iodine is deficient (Milakovic et al, 2006). In fact Nelson et al (1970) cite that the reference values of thyroid uptake need to be determined due to the geographic fluctuations and the likelihood that dietary intake in healthy person's changes.

Euthyroid Subjects

The euthyroid participants used in this study were selected from the general public, thus it is felt that this constituted a naturalistic method of determining the reference values for thyroid uptake of Technetium-99m pertechnetate in Namibia. Due to radiation safety ethical reasons all the participants were predominantly older than 40 years with an age range of 39 to 81 years. Furthermore the sample size had to be as small as possible for ethical reasons but also large enough to represent the population. Schober and Hunt (1976) articulate that a sample size of sixty is the smallest sample that would provide statistically valid results. Eighty three participants were considered for the present study and in the light of results from the serum thyroid hormone values and clinical examination 76 participants (58 females and 18 males) were selected as euthyroid. The euthyroid participants were from the Khomas region area (where there are no reports of iodine deficiency), thus they might not be considered representative of the whole of Namibia.

Thyroid Serum Hormone Value

Generally thyroid serum hormone values are used to evaluate thyroid function and to diagnose and aid in determining the cause of a thyroid disease. In this study the biochemical tests (TSH, T3 and T4) were used to determine the research participants euthyroid state. The normal range for TSH (Namibian Institute of Pathology, NIP- *unpublished*) is 0.34-5.60µIU/mL. Low TSH values confirm the diagnosis of hyperthyroidism, while high TSH values confirm the diagnosis of hypothyroidism. Thyroxine(T4) is known as the principal thyroid hormone and subsists in two forms, T4 bound to proteins, which is stored in the blood until the body needs it and the "free" T4 (fT4, tested in this study) which is the active form and is found in body tissues. The normal range for the fT4 (NIP) is 7.2 – 21.1 pmol/L. Triiodothyronine(T3) thyroid hormone is more active that the T4, and similarly to the T4 is present in either a "free" state or bound state. In some cases of hyperthyroidism the "free" T4 is normal and the "free" T3 elevated. The normal range for "free" T3 (NIP) is 3.80 - 6.00 pmol/L. (U.S. Department of Health and Human Services, National Institutes of Health, National Institute of Diabetes and Digestive and Kidney Diseases, 2010).

Figure 5.1 is an image of an incidental finding in this study. Participants with incidental findings were referred for medical management. The scintigraphic image shows no uptake of radioactivity in the thyroid, compared to the normal uptake in **Figure 5.2:**

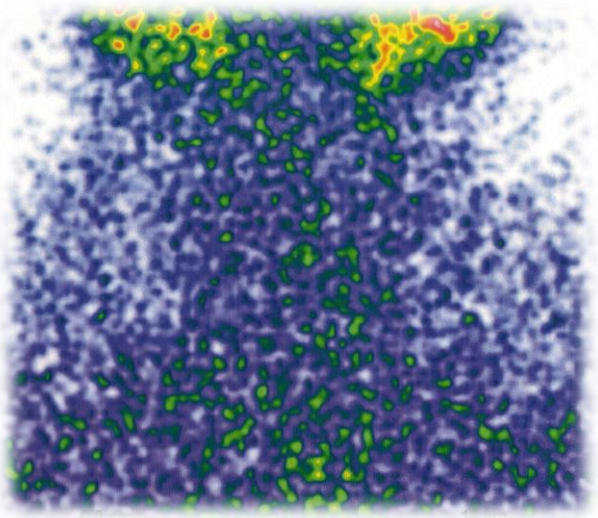

Figure 5.1 Thyroid scintigraphic image with no uptake in the thyroid.

(Image taken at research site, 2011)

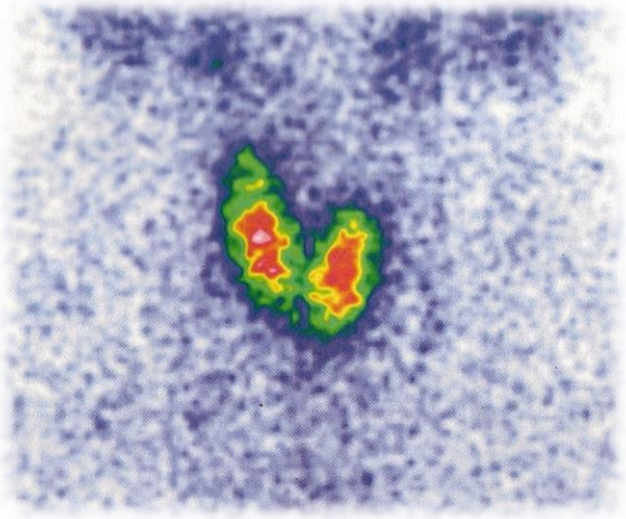

Figure 5.2 Normal Thyroid scintigraphic image

(Image taken at research site, 2011)

The blood results for **Figure 5.1** are TSH> 100, T4 - 0.06 and T3 - 2.88, Biochemically this patient is Hypothyroid and the scintigraphic findings (0.05 % uptake ofTechnetium-99 m pertechnetate) support this. **Figure 5.3** illustrates another incidental finding of a thyroid with a “Hot “nodule found from this study. The blood results for **Figure 5.3** are TSH, 4.73, T3 5.11,T4 6.51 and 2.4 %uptake of Technetium-99 m pertechnetate.

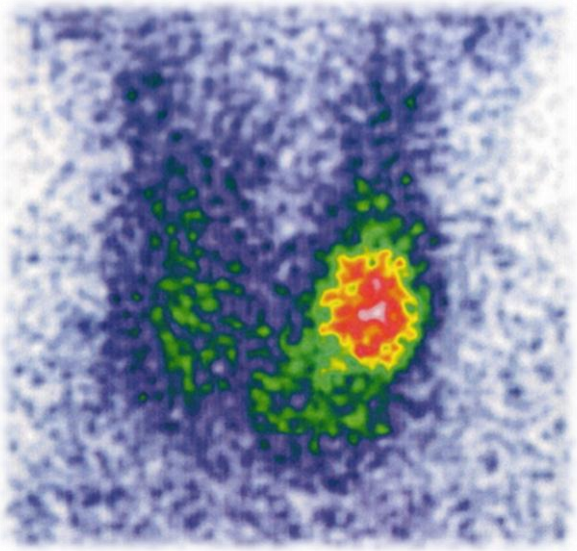

Figure 5.3 Incidental finding of a thyroid with a "Hot" nodule

(Image taken at research site, 2011)

All the 76 results used in this study for statistical analysis were of participants who had thyroid serum hormone levels (T3, T4 and TSH) within the normal laboratory reference values. All the biochemical tests were performed at the Namibian Institute of Pathology.

Radiopharmaceuticals

Iodine-131 and Iodine-123 are common radionuclides used for thyroid uptake quantification. Radioactive iodine is not used in Namibia as ^{131}I results in poor quality images, and ^{123}I is expensive and production in a cyclotron complex, also it is not readily available. Instead Technetium-99m pertechnetate is the tracer of choice used in Namibia for thyroid scanning and thyroid uptake quantification because it has a shorter half-life(6 hours), which results in a lower radiation absorbed dose to the patient. It is readily available from a Molybdenum-99 generator, and is less expensive and provides better image quality.

The administered dose of Technetium-99 m pertechnetate for thyroid uptake quantification at the research site, is 300MBq and is within the recommended dose range given in the Society of Nuclear Medicine Guidelines (74 – 370 MBq).The administered dose used in the present study was 100MBq and is within the recommended dose for thyroid uptake measurement . Calculations for adult radiation dosimetry appearing in The Society of Nuclear Medicine Guidelines for Thyroid Measurement (2006), gives the effective dose equivalent when using 74-370 MBq Technetium-99 m pertechnetate as 0.013mSv. The dose used in the present study is within the permitted ethical research categories, doses given by the Medical Research Guidelines, (2002) on ethics for medical research (Use of Biohazards and radiation).The study falls under the first category which states that the maximum permissible dose received by a research participant should not exceed 0.5 mSv.

Instrumentation

Although the thyroid probe was used by previous researchers to determine the thyroid uptake values, it was not used in the present study. Khandelwal et al, (2012) advised that Technetium-99m pertechnetate using a pinhole collimator is preferable to radioiodine as a higher radioactive dose may be administered allowing for better image resolution. In this study a NuclineTM – Spirit DH-V Dual-Head digital gamma camera with low energy high resolution (LEHR) collimator was used to perform the thyroid uptake studies on research subjects.

Comparison with Similar Studies

Table 5.1 illustrates the results of the Technetium-99m pertechnetate uptake in this study compared with the results reported from Brazil by Ramos et al (2002) who similarly evaluated normal values using Technetium-99m pertechnetate. The normal reference values obtained in Brazil were 0.4% to 1.7% of Technetium-99m pertechnetate uptake at 20 minutes which similarly was lower from the range which had previously been used (i.e. 0.3% - 3%)

The comparison of the old and new normal ranges with Namibian subjects reveals a much lower range and is consistent with the study done by Ramos et al (2002) for subjects in Brazil. The mean Technetium-99m pertechnetate uptake plus or minus two standard deviations provided the data for establishing the new normal values. As shown there was a decrease in the normal uptake values observed in both studies.

Table 5.1 Comparison of the normal thyroid uptake values between Brazil and Namibia

Group	Formula	Total	New Range	Old Range
Namibia Study	Parametric values corrected for skewness 5^{th} and 95^{th} percentile values	76	0.35% - 1.22% 0.17% - 1.7%	0.75% – 4%
Brazil	Mean +/- 2 SD	47	0.4% - 1.7%	0.3% - 3%

There was a marked difference in the traditional international normal range used in the department and the normal range found in our euthyroid participants. The traditional normal values being 0.75% -4% and the values yielded in the two methods to determine the normal values in the present study are 0.33% - 1.23% and 0.17% -1.7%. These results are consistent with those obtained in the study from Brazil. It was not possible to see if there was a statistical significance between the old and the new values as the mean for the traditional normal values is not known. Some researchers have incriminated the popularity of medications containing supplements for the lowering in normal values however in this study all vitamins and minerals taken by participants did not contain high levels of iodine (i.e. more than the recommended 0.25 mg per day). The values of Technetium-99m pertechnetate uptake observed a downward deviation from the normal values into levels previously regarded as hypothyroid. The causes of the downward deviation of normal values are not determined conclusively, but according to Anderson and Powsner (1996) the major reason

for the decrease in normal euthyroid values has been attributed to the raise in iodine ingestion.

It has been found in this study that the gender factor affects the normal value parameter slightly .The values were lower in males than in females with no statistically significant difference. According to Culp and Huskison (1978) who determined the reference values in 53 subjects together with other researchers observed a statistically significant difference in males and females, however in a study (Goldberg et al, 1964) done with a larger sample sizes this trend was not demonstrated clearly. Other studies (Gonzalez et al, 2008; Kauth et al 1972) presented a difference between males and females; it was not to a point that could be classified as statistically significant. 48.7% of the participants had uptake values below the lower limit of normal previously used in the department of 0.75% uptake at 20 minutes.

Non-Gaussian Distribution

A non-Gaussian distribution of normal values is seen, which is also noted in other literature (Ramos et al, 2002; Kulp and Huskison, 1978; Greenspan 1970; Harvey et al 1972; Nelson et al 1970). The thyroid uptake values of Technetium-99m pertechnetate range from 0.04% to 2.14 % with a mean of 0.78 %. These uptake values did not conform to a normal Gaussian distribution.
The skewed distribution illustrated here has also been reported in older studies by Harvey et al (1972), Pitman et al (1969), and Nelson et al (1969). Nelson et al (1969) gave a possible reason to the skewness to be due to including participants of different ages, and both genders in the same group as these factors are known to influence radioiodine uptake by the thyroid. Schober and Hunt (1976) who used only females to determine normal values argue well that using percentages for calculations in itself results in a skewed distribution even though the percentage values used for the calculation are distributed normally.

To normalise the distribution in the present study the *statistical test for skewness and kurtosis* formula used by Nelson et al (1969) was adopted in this study.

Historic Reference Values

Historically the literature shows either an increase or decrease in thyroid uptake values. Marked lowering of normal values have been reported by different groups Bernard et al (1970) had a previous range of 5% - 30 % (6 hours) and 15- 40% (24 hours) which was lower than the new range of 4% – 15% (6 hours) and 6- 14% (24 hours) . Harvey et al (1972)

had previous values of 15% – 45% (24 hours) and new values of 0.8% -65% (24 hours), while others have reported upward deviations in their values compared to the values they had established in earlier years; Wong and Schultz, (1977) had radioactive iodine uptake values of 20.5% ±6.1%(SD) at 24 hours where for years previously it was found to be 11.5% ± 8 % (SD). In more recent literature Kusai and Tashtoush (2010) noted a lowering of normal values (15% ± 7% SD at, 24 hours) for the Jordan population which was comparable to literature by the Americans; Pitman et al (1967) who had previous uptake of 28.6% ± 6.5% in 1959 and in later years 15.4%± 8.8% in 1967-1968.Blum and Chandra (1971) had uptake values that changed from 20% – 45% (24 hours) in 1963 to 10%-34% (24 hours) in 1970. Also Milakovic et al (2006) reported that their Swedish population presented with lower values (11% - 33 % at 24 hours) than that obtained (14% - 30%at 24 hours) half a century before for that same population. Values obtained by Gonzalez et al (2008) for Chile did not differ from their previous departmental values.

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CHAPTER SIX CONCLUSION

The contribution of thyroid uptake studies in the evaluation of the thyroid gland has motivated for several studies in determining normal reference values for thyroid uptake studies, as it is an essential complementary diagnostic tool in studying the physiology of the thyroid gland. Furthermore it is used by Nuclear Medicine physicians to make a distinction between hyperthyroidism and other types of thyrotoxicosis and to determine the dose of ^{131}I for the treatment of hyperthyroidism and the ablative dose for therapy (ARC, SNM, SPR revised guidelines, 2009).
The normal reference values for thyroid uptake of Technetium-99m pertechnetate, ^{131}I and ^{123}I in euthyroid persons, changes with the geographic location, and may change in one area from one decade to the next.
This study determined the thyroid uptake reference values of Technetium-99m pertechnetate for the Namibian population. The values in this study were compared to those obtained from similar studies and the internationally known values.

This study showed that there was a marked difference in the traditional normal range used in the department and the normal range found in the euthyroid participants. It was not possible to see if there was a statistical significance between the former and the latter as the mean for the traditional normal values is not known. The traditional normal values are 0.75% - 4% and the values obtained in the study are 0.35% - 1.22% and 0.17% - 1.7% of Technetium-99m pertechnetate at 20 minutes. The deviated values are considered by the author to be due to the iodized table salt used by the population in conjunction with other foods (dairy products and sea food) containing iodine.

Limitations

The results from this study provide information on the normal reference values for thyroid uptake of Technetium-99m pertechnetate in the Namibian population. The study was however limited to the Khomas region as it was the only region in Namibia that had a Nuclear Medicine Department during the time of the research study (August 2011 to December 2011). Although the department receives referrals from all thirteen regions of Namibia, the majority of the population migrate to the Khomas region for employment. However the Ovahimba, Kavangos, Caprivians, Bushmen and Tswana ethnic groups were not included amongst the research subjects.

Another limitation in the research methodology was the small sample size (sample size 76) and the age limit of 40 years and above. A larger sample size and including participants with ages 18 to 40 would better represent the population; also there are patients who present with thyroid disorders who are of child bearing age (18 - 40 years). Further limitations include; failure to determine the level of urinary iodine

Recommendations

It is not always convenient for each Nuclear Medicine facility to determine thyroid uptake normal range in euthyroid individuals, however the iodine intake fluctuates without public consciousness, and thus the greatest pitfall in the thyroid uptake test is the omnipresent iodine atom incurred in our daily lives. For this reason, similar to other studies on thyroid uptake reference values, it is recommended that the reference ranges for thyroid uptake of Technetium-99m pertechnetate for different Nuclear Medicine facilities are reviewed.

Furthermore it is recommended that the new values obtained in this study for thyroid uptake of Technetium-99m pertechnetate be used for the Namibian population when determining thyroid uptake percentage in patients suspected of thyroid disorders, more specifically the normal range of 0.17% - 1.7 % should be adopted by the Nuclear Medicine department at the Windhoek Central Hospital. Thus the department protocol for the thyroid uptake measurements should be altered to accommodate the new reference values for thyroid uptake of Technetium-99m pertechnetate. It is also recommended that future studies should recruit a larger sample size (100 subjects. Also with ethical justification all age groups (18 years and above) can be used to better represent the whole population.

The research method used in this study is the standard protocol used in the Nuclear Medicine Department, which is adopted from the Society of Nuclear Medicine Guidelines for Thyroid Uptake Measurement. As such Nuclear Medicine Departments will be able to adopt their respective routine thyroid uptake protocols to determine their thyroid uptake reference values without clinical justification.

This study provides new evidence supporting the importance of periodic checks on the normal reference values for thyroid uptake of Technetium-99m pertechnetate.

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APPENDIX 1 RESEARCH VOLUNTEER FILE

Appendix 1A	Questionnaire
Appendix 1B	Consent Form
Appendix 1C	Data Capture Sheet
Appendix 1D	Research Methodology checklist
Appendix 1E	Blood Form
Appendix 1F	Research Notice

Appendix 1A Research Questionnaire

RESEARCH QUESTIONNAIRE

Questionnaire designed to reveal contraindications for thyroid uptake studies,iodine contamination or ingestion and diet

Volunteer code:…….

MEDICAL HISTORY

1. **Have you been diagnosed with any thyroid disorders in the past/have you been diagnosed with any thyroid disorders at present.**
   ………..
   ……..

2. **Has any immediate family member (grandparents, parent, siblings or child) ever been diagnosed with any thyroid disorders?**
   ……..
   ……..

3. **Are you on any medication/oestrogen (thyroid hormone, antithyroid drugs or medication containing iodine? (If on medication what medication)**?
   .....
   .....
   ……..

4. **Have you been exposed to iodinated contrast (CT scan etc)?**
   ……..……
   ……..

5. **In the last week, have you used any iodine-containing (brown coloured antiseptic skin cleaner?**
   ……..
   ……..

6. **In the last two years, have you taken any heart medication (medicine for abnormal heart rhythms?**

.....
.....
.....

7. **For WOMEN, in the last week have you used any iodine – containing vaginal douche?**

……
……

8. **Do you smoke cigarettes? (In the last 24 hours how many have you smoked?**

……
……

9. **For WOMEN, Are you pregnant/ nursing (when was the last menstrual period)?**

……
……
.....

10. **Have you been recently exposed to radionuclides?**

……
……

SOURCE OF IODINE IN YOUR DIET

1. **Do you use iodized salt in your food?**

……
……

2. **In the last 24 hours have you added salt to your food, (haw many tea spoons)?**

……
……

3. **In the last 24 hours have you eaten any sea food (how often do you eat sea food)**

……
……

4. **Are you taking any multivitamins (what kind , are they containing any iodine)**

……
……

5. **In the last 24 hours have you had any of the following products to eat or drink (cow's milk, cheese, yogurt, Ice cream, frozen yogurt, how many servings)?**

……
……

6. **In the last 24 hours have you eaten eggs, how many?**

……
……

7. **In the last 24 hours have you eaten bread that was not home made, how many slices?**

……
……

Volunteer accepted? YES/NO

Volunteer not accepted (reason)

……
……
……
……
……
……

Appendix 1B Research Participants Consent Form

PARTICIPANT INFORMATION AND INFORMED CONSENT FORM FOR RESEARCH PROJECT TO DETERMINE NORMAL REFERANCE VALUES FOR THYROID UPTAKE OF Technetium-99m pertechnetate

(BLOOD SAMPLE COLLECTION AND Technetium-99m pertechnetate ADMINISTRATION)

PRINCIPAL INVESTIGATOR: Roswita H Hamunyela

CAPE PENINSULA UNIVERSITY OF TECHNOLOGY

Cape Peninsula University of Technology

Health & Wellness Sciences
Radiography
Dep.of Nuclear Medicine
Grote Schuur Hospital

NOTE: This consent form consists of 4 Sections:

1) **SECTION 1, General information for All research participants(Patient & Non Patient volunteers)**
2) **SECTION 2, for participants40 years and above ONLY (Permission to perform thyroid uptake study).**
3) **SECTION 3, for patient participants ONLY (Permission to use results from thyroid uptake study)**
4) **SECTION 4, Declaration by Volunteer, Principal investigator and Translator.**

SECTION 1

General information for ALL research volunteers
(Patient & Non Patient volunteers

As Nuclear Medicine technologists it is part of our goal to contribute to continuously offer quality medical care to our clients, as well as continuously striving into research ventures that will bring new knowledge, enabling us to improve our patient management services,
On this note we request your participation in a hospital/clinic project that strives to determine reference values for thyroid uptake of Technetium-99m pertechnetate, for the Namibian population.

Please take some time to read the information presented here which will explain the details of this project. Please ask the principal investigator about any part of the project that you do not fully understand. It is very important that you are fully satisfied as to what the research entails. Your participation is **entirely voluntary** and you are free to withdraw at any stage if you say no, this will not affect you negatively in any way whatsoever.

The research study has been approved by the Permanent Secretary (Ministry of health and social services.

PURPOSE OF RESEARCH STUDY

To investigate thyroid uptake of 99mTc-pertechnetate in healthy subjects in order to develop potential reference values for the Namibian population.

BENEFITS

Department benefits:

1. Development of possible departmental normal reference values for 99mTc-pertechnetate thyroid uptake studies for the Namibian population.
2. Comparison of the normal reference values with international normal reference values.

3. Improved accuracy /validity of the Thyroid uptake Study done in the Nuclear Medicine Department at the Dr B May Cancer Care Centre
4. Improved management of patients referred to the Dr B May Cancer Care Centre's Nuclear Medicine Department with suspected thyroid disorders.

Volunteer Benefits:

Are there any benefits (financial/nonfinancial) to your taking part in this study?

There are no direct benefits to you as a volunteer. The study will help us determine the thyroid uptake reference values for the Namibian population which will help the department of nuclear medicine to better manage patients referred for thyroid disorders.

You will not be paid to take part in this study. In the unlikely event that the research leads to the development of a commercial application, you or your family will not receive any profits or royalties. However profits will be reinvestigated to support the cause of future research, which may bring benefits to your family or community in future.

There will be no gifts, or any extra course credit.

Investigators Benefits:

Will the investigator benefit (financially/nonfinancial) from this research?

The researcher will not benefit financially from the study

SECTION 2

Participants40 years and above

(Permission to perform thyroid uptake study).

INFORMATION ABOUT STUDY

Why have you been invited to participate?

I am looking for healthy subjects who are not on medication, have no presence of thyroid disorders and have normal thyroid function blood results.

What does this research study involve?

The study involves participantsundergoing the following:

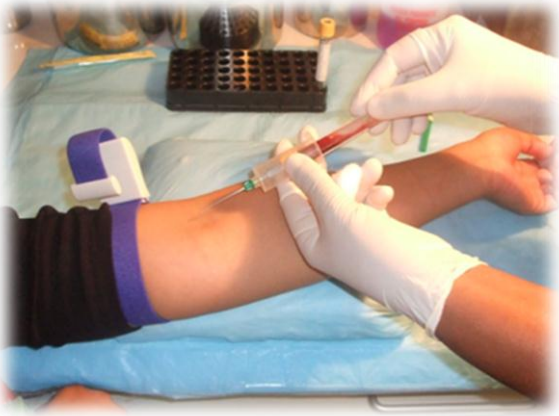
1. ***Answering a Questionnaire***

The questionnaire is designed to identify any contraindications or any form of iodine contamination that could interfere with the thyroid uptake study.

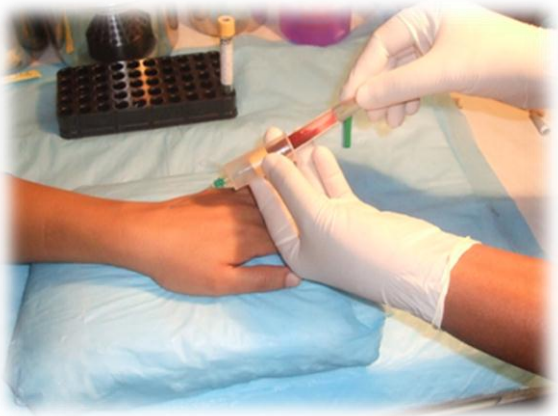
2. ***Collection of blood sample***

A blood sample of 4ml needs to be taken from each participating volunteer to test the thyroid hormone blood values of the TSH T3 & T4.

The blood will be taken using a 5ml syringe, and will be withdrawn from the inside of the elbow) or left or right hand. The blood will be taken once only for the study.



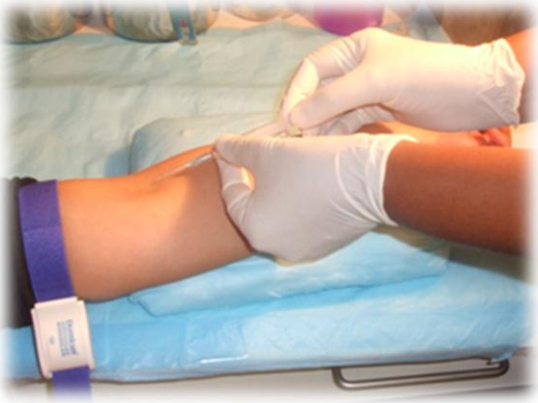
Arm



or hand

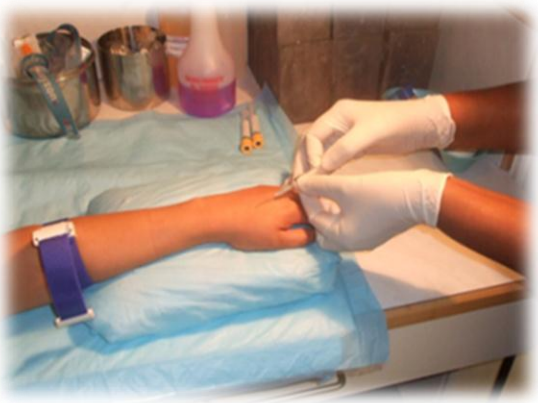
3. Injection of radioactivity

A thyroid uptake study involves being injected with 100MBq (Technetium-99m pertechnetate) radioactive liquid which is made up to a volume of less than 2ml. The amount of radioactivity given is within the recommended dose limit (37-370MBq). The injection is given with a 2ml syringe on the inside of the elbow or left or right hand. The injection will be given once only



Arm injection

or



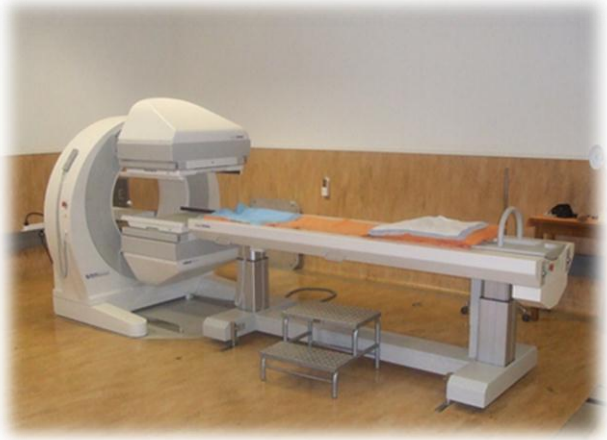
hand injection

4. Imaging the thyroid

Twenty minutes after injecting the radioactive solution a thyroid uptake scan will be done.

You will be placed on the gamma camera bed on your back, with a pillow under your shoulders, the face of the camera will be 20cm over the your neck and the thyroid image will be obtained.

Gamma Camera used for thyroid Imaging



RADIATION PROTECTION

Participantswill not be subject to other unnecessary radiation exposure.

RISKS, STRESS OR DISCOMFORTS

Radiation risk

The radiation risk will be minimal and is within the acceptable limits. The estimated dose you will receive is 9mSv.

(The recommended annual dose limit for general public may be greater than 5mSV but less that 20mSV (ICRP – International Commission on Radiological Protection).

Adverse reactions:

There are no known adverse side effects to this procedure.

Questionnaire:

The questionnaire you will be asked to complete does not disclose any sensitive information or cause any psychological discomfort.

In an event where the information on the questionnaire shows that you cannot volunteer, this does not mean that you have a disease of the thyroid. Being excluded from the study would be for the following reasons:

1. Iodine in the food you have eaten that will affect the study.
2. Allergy to iodine.
3. Suspected pregnancy.

Blood sample

There are no risks to you when providing a blood sample for this study.

Equipment

The gamma camera being used is used for every day imaging and will not cause you any harm.

CONFIDENTIALITY

How will your confidentiality be protected?

All the data being collected in this research will be coded, and your name will not be used to ensure confidentiality. The data will be stored in the department safe, and only the investigator, physician and registered nurse will handle the data.

DATA BEING COLLECTED

The following data will be collected from each volunteer:

- Age
- Gender (male/female)
- TSH blood levels
- T3 blood levels
- T4 blood levels
- Percentage thyroid uptake

No other data will be collected.

The Blood levels for the thyroid function tests are used to evaluate the thyroid's functioning, and will be used in this study to see if your thyroid is functioning normally.

SECTION 3

Patient volunteers

(Permission to use results from thyroid uptake study)

With regards to the research study described above the research investigator would here with like to use the following data from your diagnostic thyroid uptake study:

- Age
- Gender (male/female)
- TSH blood levels
- T3 blood levels
- T4 blood levels
- Percentage thyroid uptake

No other data will be collected.

Why have you been invited to participate?

I am looking for healthy patients who have been referred to the Nuclear Medicine department, who have no presence of thyroid disorders and have normal thyroid function and blood results.

How will your confidentiality be protected?

All the data being collected in this research will be coded, and your name will not be used to ensure confidentiality. The data will be stored in the department safe, and only the investigator, physician and registered nurse.

NOTE: FOR ALL PARTICIPANTSNB….!!!

In an event where you are found to have abnormal blood results, the Nuclear Medicine physician at the Dr B May cancer care center will call you in for counseling, giving a clear explanation of the blood results, and you will be given an option of being referred to a state doctor at the hospital or your own private doctor who will decide on further patient management. Medical costs to private doctors will be deducted from volunteer's medical aid, and all referrals to state doctors cost N$ 15, 00 for consultation.

SECTION 4,

(Declaration by Volunteer, Principal investigator and Translator.)

Declaration by volunteer:

By signing below, I…….agree to take part in a research study to determine reference values for thyroid uptake of Technetium-99m pertechnetate .

I declare that:

- I have read or had read to me this information and consent form and it is written in a language with which I am fluent and comfortable. I have had a chance to ask questions and all my questions have been adequately answered
- I understand that taking part in this study is voluntary and I have not been pressurized to take part.
- I understand that all data collected in this research will be handled confidentially.

Signed at (place)……on (date)……

…… ……

Signature of participant Signature of witness

Declaration by principal investigator:

I (name) ……declare that:

- I explained the information in this document to …….
- I encouraged him/her to ask questions and took adequate time to answer them.
- I am satisfied that he/she adequately understands all aspects of the research as discussed above.
- I did/did not use a translator.

Signed at (place)……on (date)……

…… ……

Signature of investigator Signature of witness

Declaration by language translator

I (name) ……declare that:

- I assisted the investigator (name) ……..to explain the

  Information in this document to (name of participant) ……..using the medium of ……...

- We encouraged him/her to ask questions and took adequate time to answer them.
- I conveyed a factually correct version of what was related to me.
- I am satisfied that the participant fully understands the content of this informed consent document and has had all his/her question satisfactorily answered.

Signed at (place)……on (date)……

…… …….

Signature of translator Signature of witness

_____

Appendix 1C Data Capture Sheet

Data capture sheet for each volunteer

Note: Type of volunteer mark with (X)

1. Patient with normal thyroid functions…….
2. Volunteer over 40 years……..

Ethnicity: ***please circle appropriate ethnic group***

Herero, Ovahimba, Kavango, Caprivian, Damaras, Namas, Bushmen and Coloured, Caucasians, Other…….

Date……

Confidentiality code: ……

Gender: Male/Female: ……

Age: ……

Blood variables

THYROID HORMONE	Normal range	Volunteer sample value
TSH		
T3		
T4		

Normal Technetium-99m pertechnetateThyroid uptake values (%) ……

% Thyroid uptake value (Volunteer) ……

Appendix 1D Research Methodology Check List

Research Methodology Checklist (to be filled in for each volunteer)

✓ **Tick off once done.** **Volunteer code:……**

-

Procedure explained to volunteer	
Consent form signed	
Blood drawn	
Full syringed measured (Dose…….....)	
Radioactivity injected:	
Right cubital fossa	
Left cubital fossa	
Right hand	
Left hand	
Empty syringe measured	
Questionnaire completed	
Blood variables collected from lab	
%Thyroid uptake recorded	

Radioactivity injected & Blood drawn by:

Qualified Nurse: Name…… Signature……

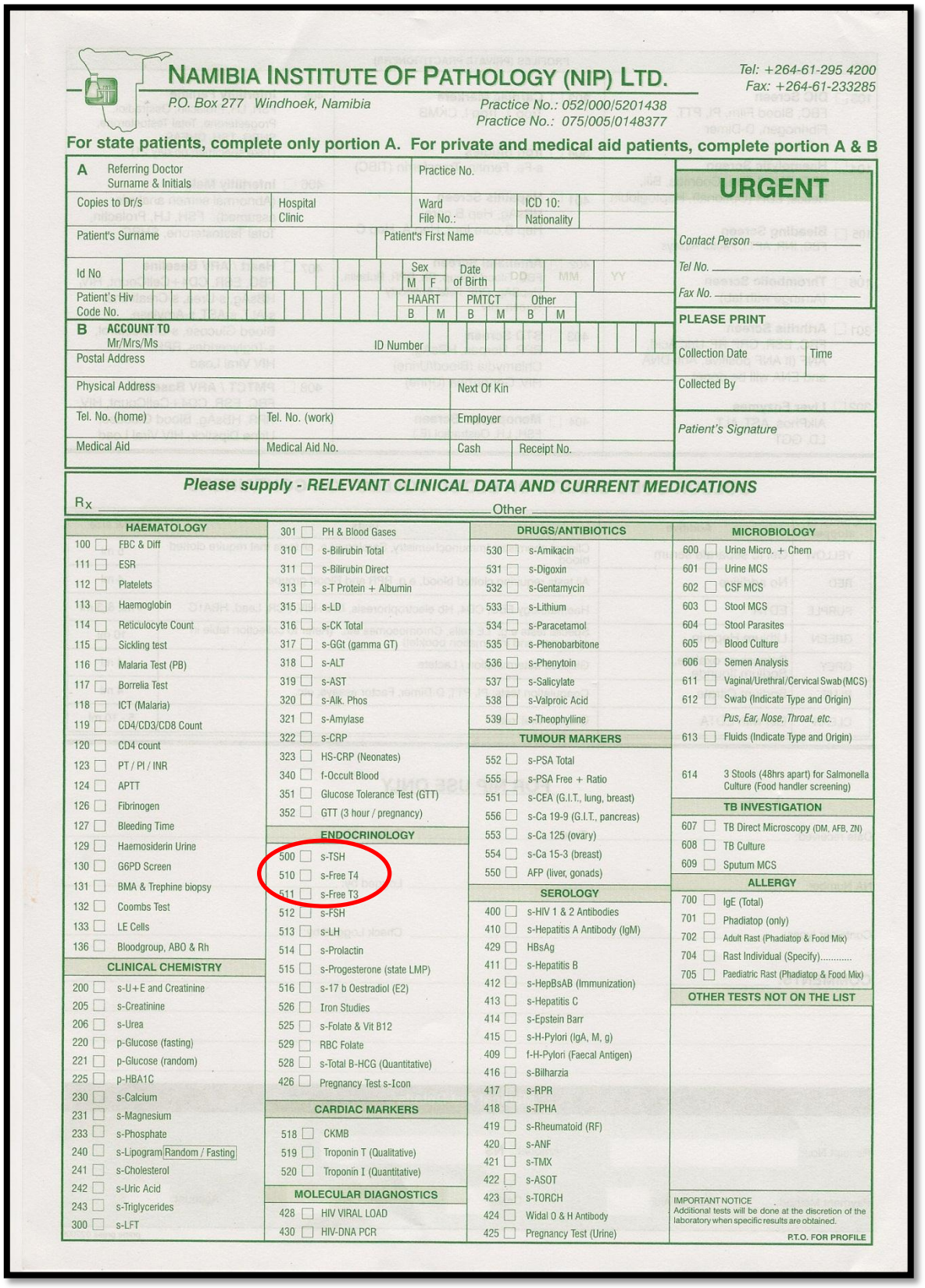
Thyroid uptake scan performed by:

Qualified Technologist: Name……... Signature……....

Thyroid uptake processed by:

Qualified Doctor: Name……. Signature…….

Appendix 1E Blood Form



NAMIBIA INSTITUTE OF PATHOLOGY (NIP) LTD.

P.O. Box 277 Windhoek, Namibia

Practice No.: 052/000/5201438
Practice No.: 075/005/0148377

Tel: +264-61-295 4200
Fax: +264-61-233285

For state patients, complete only portion A. For private and medical aid patients, complete portion A & B

A Referring Doctor Surname & Initials | Practice No.

Copies to Dr/s | Hospital Clinic | Ward File No.: | ICD 10: | Nationality

Patient's Surname | Patient's First Name

Id No | Sex M F | Date of Birth DD MM YY

Patient's Hiv Code No. | HAART B M | PMTCT B M | Other B M

B ACCOUNT TO Mr/Mrs/Ms | ID Number

Postal Address

Physical Address | Next Of Kin

Tel. No. (home) | Tel. No. (work) | Employer

Medical Aid | Medical Aid No. | Cash | Receipt No.

URGENT

Contact Person _____
Tel No. _____
Fax No. _____

PLEASE PRINT

Collection Date | Time

Collected By

Patient's Signature

Please supply - RELEVANT CLINICAL DATA AND CURRENT MEDICATIONS

Rx _____ Other _____

HAEMATOLOGY

- 100 FBC & Diff
- 111 ESR
- 112 Platelets
- 113 Haemoglobin
- 114 Reticulocyte Count
- 115 Sickling test
- 116 Malaria Test (PB)
- 117 Borrelia Test
- 118 ICT (Malaria)
- 119 CD4/CD3/CD8 Count
- 120 CD4 count
- 123 PT / PI / INR
- 124 APTT
- 126 Fibrinogen
- 127 Bleeding Time
- 129 Haemosiderin Urine
- 130 G6PD Screen
- 131 BMA & Trephine biopsy
- 132 Coombs Test
- 133 LE Cells
- 136 Bloodgroup, ABO & Rh

CLINICAL CHEMISTRY

- 200 s-U+E and Creatinine
- 205 s-Creatinine
- 206 s-Urea
- 220 p-Glucose (fasting)
- 221 p-Glucose (random)
- 225 p-HBA1C
- 230 s-Calcium
- 231 s-Magnesium
- 233 s-Phosphate
- 240 s-Lipogram Random / Fasting
- 241 s-Cholesterol
- 242 s-Uric Acid
- 243 s-Triglycerides
- 300 s-LFT
- 301 PH & Blood Gases
- 310 s-Bilirubin Total
- 311 s-Bilirubin Direct
- 313 s-T Protein + Albumin
- 315 s-LD
- 316 s-CK Total
- 317 s-GGt (gamma GT)
- 318 s-ALT
- 319 s-AST
- 320 s-Alk. Phos
- 321 s-Amylase
- 322 s-CRP
- 323 HS-CRP (Neonates)
- 340 f-Occult Blood
- 351 Glucose Tolerance Test (GTT)
- 352 GTT (3 hour / pregnancy)

ENDOCRINOLOGY

- 500 s-TSH
- 510 s-Free T4
- 511 s-Free T3
- 512 s-FSH
- 513 s-LH
- 514 s-Prolactin
- 515 s-Progesterone (state LMP)
- 516 s-17 b Oestradiol (E2)
- 526 Iron Studies
- 525 s-Folate & Vit B12
- 529 RBC Folate
- 528 s-Total B-HCG (Quantitative)
- 426 Pregnancy Test s-Icon

CARDIAC MARKERS

- 518 CKMB
- 519 Troponin T (Qualitative)
- 520 Troponin I (Quantitative)

MOLECULAR DIAGNOSTICS

- 428 HIV VIRAL LOAD
- 430 HIV-DNA PCR

DRUGS/ANTIBIOTICS

- 530 s-Amikacin
- 531 s-Digoxin
- 532 s-Gentamycin
- 533 s-Lithium
- 534 s-Paracetamol
- 535 s-Phenobarbitone
- 536 s-Phenytoin
- 537 s-Salicylate
- 538 s-Valproic Acid
- 539 s-Theophylline

TUMOUR MARKERS

- 552 s-PSA Total
- 555 s-PSA Free + Ratio
- 551 s-CEA (G.I.T., lung, breast)
- 556 s-Ca 19-9 (G.I.T., pancreas)
- 553 s-Ca 125 (ovary)
- 554 s-Ca 15-3 (breast)
- 550 AFP (liver, gonads)

SEROLOGY

- 400 s-HIV 1 & 2 Antibodies
- 410 s-Hepatitis A Antibody (IgM)
- 429 HBsAg
- 411 s-Hepatitis B
- 412 s-HepBsAB (Immunization)
- 413 s-Hepatitis C
- 414 s-Epstein Barr
- 415 s-H-Pylori (IgA, M, g)
- 409 f-H-Pylori (Faecal Antigen)
- 416 s-Bilharzia
- 417 s-RPR
- 418 s-TPHA
- 419 s-Rheumatoid (RF)
- 420 s-ANF
- 421 s-TMX
- 422 s-ASOT
- 423 s-TORCH
- 424 Widal O & H Antibody
- 425 Pregnancy Test (Urine)

MICROBIOLOGY

- 600 Urine Micro. + Chem
- 601 Urine MCS
- 602 CSF MCS
- 603 Stool MCS
- 604 Stool Parasites
- 605 Blood Culture
- 606 Semen Analysis
- 611 Vaginal/Urethral/Cervical Swab (MCS)
- 612 Swab (Indicate Type and Origin) *Pus, Ear, Nose, Throat, etc.*
- 613 Fluids (Indicate Type and Origin)
- 614 3 Stools (48hrs apart) for Salmonella Culture (Food handler screening)

TB INVESTIGATION

- 607 TB Direct Microscopy (DM, AFB, ZN)
- 608 TB Culture
- 609 Sputum MCS

ALLERGY

- 700 IgE (Total)
- 701 Phadiatop (only)
- 702 Adult Rast (Phadiatop & Food Mix)
- 704 Rast Individual (Specify).....
- 705 Paediatric Rast (Phadiatop & Food Mix)

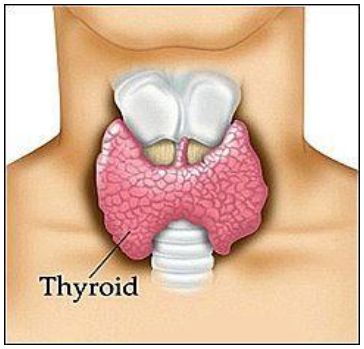
OTHER TESTS NOT ON THE LIST

IMPORTANT NOTICE
Additional tests will be done at the discretion of the laboratory when specific results are obtained.

P.T.O. FOR PROFILE

Appendix 1F Research Notice

Participants*required to participate in aNuclear Medicine study*



Establishment of normal reference values for Technetium-99m pertechnetateThyroid uptake studies for the Namibian population

We would like to invite participantsto participate in a Nuclear Medicine Research study. At the Dr Bernard May Cancer Centre (Windhoek Central Hospital) we are establishing normal reference values for Technetium-99m pertechnetatethyroid uptake studies, for the Namibian population.

Normal reference values for thyroid uptake studies are necessary as they determine whether or not a patient has a thyroid disease.

The aim of the study is to establish normal reference ranges for Technetium-99m pertechnetate thyroid uptake studies, for the Namibian population, to help the Nuclear Medicine department improve patient management.

For this investigation we need healthy participantswho are not of child bearing age (age 40 years and above) who do not take any medication, to undergo the routine thyroid uptake scan, which requires imaging with radioactive Technetium-99m pertechnetate, and a blood sample to determine the thyroid functions.

The study is purely a diagnostic procedure, the radioactive Technetium-99m pertechnetate used in this study is used in the Nuclear Medicine department daily to perform all diagnostic imaging studies, apart from the blood sample nothing is taken from the volunteer, no medication is given to the volunteer either. Withdrawing such a small volume of blood (4ml) causes no harm. The radioactivity (100MBq) being used has no side effects.
The thyroid uptake will be performed with a Nuclear Medicine gamma camera.

NOTE: There are NO benefits financially in taking part in this research study:
Healthy volunteer may however unexpectedly become a direct or indirect beneficiary of this research if it brings new knowledge, thus the acquisition of knowledge may be of no immediate benefit to the healthy volunteer.

Duration of study: Aug 2011- December 2011

*If you are willing to participate in this study please sms ,**NAME ,VOLUNTEER , CONTACT**NO to:*

R H Hamunyela
Nuclear Medicine Technologist
Dr Bernard May Cancer Care Centre (Windhoek Central Hospital
Tel: 0814176692
Work: 061 2033264
roswitahamunyela@hotmail.com

APPENDIX 2 HELSINKI DECLARATION

APPENDIX 2 HELSINKI DECLARATION

HELSINKI DECLARATION

World Medical Association Declaration of Helsinki

ICH Guideline for Good Clinical Practice (GCP)

- Clinical trials should be conducted in accordance with the ethical principles that have their origin in

- Declaration of Helsinki and that are consistent with GCP and the applicable regulatory requirement(s).

- Before a trial is initiated, foreseeable risk and inconveniences should be weighed against the anticipated benefit for the individual trial subject and society. A trial should be initiated and continued if the anticipated benefits justify the risk.

- The rights, safety and wellbeing of the trial participants are the most important considerations and should prevail over interest of science and society.

- The available non-clinical and clinical information on an investigational product should be adequate to support the proposed clinical trials.

- Clinical trials should be scientifically sound, and described in a clear, detailed protocol.

- A trial should be conducted in compliance with the protocol that has received prior institutional review board (IRB)/independent ethics committee (IEC) approval/ favorable opinion.

- The medical care given to, and medical decisions made on behalf of, participants should always be the responsibility of the qualified physician or, when appropriate, of a qualified dentist.

- Each individual involved in conducting a trial should be qualified by education, training, and experience to perform his or her respective task(s).

- Freely given informed consent should be obtained from every participant prior to clinical trial participation.

- All clinical trial information should be recorded, handled, and stored in a way that allows its accurate reporting, interpretation and verification.

- The confidentiality of records that could identify participants should be protected, respecting the privacy and confidentiality rules in accordance with the applicable regulatory requirement(s).

- Investigational product should be manufactured, handled, and stored in accordance with applicable good manufacturing practice (GMP). They should be used in accordance with the approved protocol.

- Systems with procedures that assure the quality of every aspect of the trial should be implemented.

APPENDIX 3 ETHICS APPROVALS

Appendix 3A Ethics Approval (Cape Peninsula University of Technology

Appendix 3B Ethics Approval (Ministry of Health and Social Services –Namibia)

Appendix 3C Ethics Approval (Namibian Institute of Pathology)

Appendix 3D Ethics Approval (National Radiation Protection Authority- Namibia

Appendix 3A Ethics Approval (Cape Peninsula University of Technology)

Cape Peninsula
University of Technology

25 July 2011
CPUT/HW-REC 2011/H14

P.O. Box 1906 • Bellville 7535 South Africa •Tel: +27 21 442 6162 • Fax +27 21 447 2963
Symphony Road Bellville 7535

OFFICE OF THE CHAIRPERSON:
HEALTH AND WELLNESS SCIENCES RESEARCH ETHICS COMMITTEE (HW-REC)
Registration Number NHREC: REC- 230408-014

At the meeting of the Health and Wellness Sciences-REC on 22 June 2011 approval was granted to Roswita Hamunyela pending amendments that have now been received and reviewed. This approval is for research activities related to an MTech: Radiography at this institution.

TITLE:

Establishment of normal reference values of thyroid uptake of Technetium-99m pertechnetate for the Namibian population

INTERNAL SUPERVISOR: **Ms G Philptheou**

EXTERNAL CO-SUPERVISOR: **Dr T Kotze**

Comment:

Research activities are restricted to those detailed in the revised proposal and application submitted in June 2011

Approval will not extend beyond 25 July 2012. An extension must be applied for should data collection for this study continue beyond this date.

Prof PENELOPE ENGEL-HILLS
CHAIR: HEALTH AND WELLNESS SCIENCES RESEARCH ETHICS COMMITTEE

e-mail: *engelhillsp@cput.ac.za*

Appendix 3B Permission– (Ministry of Health and Social Services)

9 - 0/0001

REPUBLIC OF NAMIBIA

Ministry of Health and Social Services

Private Bag 13198 | **Ministerial Building** | **Tel: (061) 2032510**
Windhoek | **Harvey Street** | **Fax: (061) 227786**
Namibia | **Windhoek** | **E-mail: eshaama@mhss.gov.na**
Enquiries: Ms. E.N. Shaama | **Ref.:** 17/3/3 | **Date:** 30 May 2011

OFFICE OF THE PERMANENT SECRETARY

Ms. Roswita H. Hamunyela
P.O. Box 24437
Windhoek

Dear Ms. Hamunyela

Re: Establishing normal references values for thyroid uptake of Technetium-99m Pertechnetate in Namibian population

1. Reference is made to your application to conduct the above-mentioned study.

2. The proposal has been evaluated and found to have merit.

3. Kindly be informed that permission to conduct the study has been granted under the following conditions:

3.1 The data to be collected must only be used for completion of your Masters in Technology-Nuclear Medicine Degree;
3.2 No other data should be collected other than the data stated in the proposal;
3.3 A quarterly report to be submitted to the Ministry's Research Unit;
3.4 Preliminary findings to be submitted upon completion of study;
3.5 Final report to be submitted upon completion of the study;
3.6 Separate permission should be sought from the Ministry for the publication of the findings.

Yours sincerely,

MR. K. KAHUURE
PERMANENT SECRETARY

"Health for All"

Appendix 3C Approval–(Namibian Institute of Pathology)

NAMIBIA INSTITUTE OF PATHOLOGY (NIP) LTD.

Tel: +264-61-295 4200, Fax: +264-61-255 566, P.O. Box 277, Windhoek, Namibia

Reg. No. 2000/431

OFFICE OF THE CHIEF EXECUTIVE OFFICER

Enquiries: H.T Kaura

01 August 2011

Ms. Roswita H. Amunyela
P O Box 24437
Windhoek

RE: ESTABLISHING NORMAL REFRENCE VALUES FOR THYROID UPTAKE OF TECHNETRUM-99M PERTECHNELATE IN NAMIBIA POPULATION

1. Reference is made to your application to conduct the above mentioned study.

2. The proposal has been evaluated and granted approval under the following conditions:

   a) The cost of the analysis (blood tests on thyroid functions (TSH, T3 and T4) will be covered by the Windhoek Central Hospital (WCH).
   b) Arrangement will be made with NIP to ensure that the billing for these tests are invoiced to the WCH.

Yours sincerely

NAMIBIA INSTITUTE OF PATHOLOGY (NIP) LTD
2011 -08- 02
P. O. Box 277 WINDHOEK NAMIBIA

Mrs. TK Angula
Chief Executive Officer

Appendix 3D Ethics Approval – (National Radiation Protection Authority)

9 - 0/0001

REPUBLIC OF NAMIBIA

Ministry of Health and Social Services

P/BAG 13198
Windhoek
Namibia
Enquiries: *Mr. P. Ngalangi*

Ministerial Building
Harvey Street
Windhoek
Ref/8/04/12/019_11/02

Tel: 203 2414/18
Fax: 234 083
email: vuushona@yahoo.com
Date: 31 August 2011

NATIONAL RADIATION PROTECTION AUTHORITY

Windhoek Central Hospital-Radiation Oncology
P/Bag 13215
Windhoek

Attention: Ms. R. Hamunyela

Subject: **Research on "*Establishing Normal Reference Values for Thyroid Uptake of Technetium-99m Pertechnetate for the Namibian Population*".**

The above subject matter has reference.

The NRPA concurs with the proposed research on the basis that the outcome of the research will contribute to an enhanced diagnostic protocol for abnormal thyroid conditions. Therefore there is justification as it apears in the submission that there will be notable benefit to the population at large.

While concurring with the proposal, the NRPA is also mindful of the radiological risk involved. Therefore it is directed that the dose to the cohort must be optimised. In this respect the number of cohorts must be kept to a minimum and the dose to be administered must be minimum practical.

Furthermore the research must be conducted within the framework of the provisions of the Helsinki Declaration and the recommendations of the International Commission for Radiation Protection (ICRP) (*ICPP Publication 103*).

Sincerely Yours,

Mr. A. Tjiparuro
Director

"Health for All"