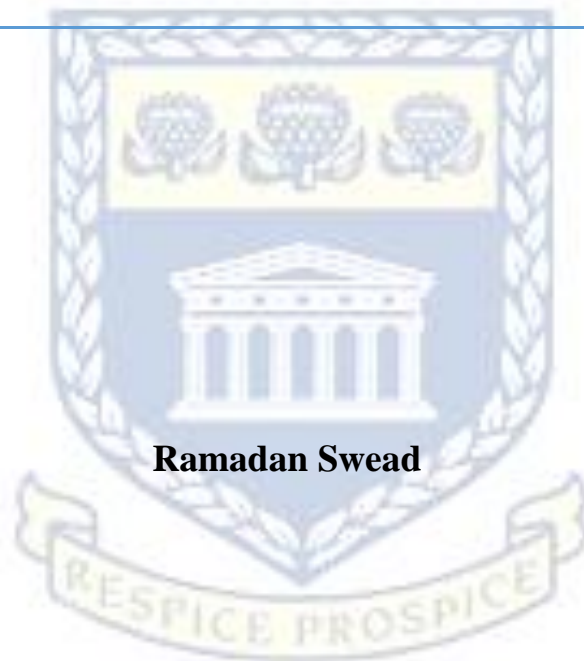


The development of a study protocol, and ethics and regulatory approval documentation, for evaluation of clinical efficacy of *Sutherlandia frutescens* in adult type-2 diabetics



Ramadan Swead

A thesis submitted in fulfilment of the requirements for the degree of Magister Scientiae (Pharmaceutical Sciences) in the Discipline of Pharmacology at the University of the Western Cape, Bellville, South Africa.

Supervisor: Professor James A Syce

The development of a study protocol, and ethics and regulatory approval documentation, for evaluation of clinical efficacy of *Sutherlandia frutescens* in adult type-2 diabetics

The logo of the University of the Western Cape is a shield-shaped crest. The top section is yellow with three blue floral motifs. The middle section is blue with a white classical building facade. The bottom section is blue with a white banner that reads "RESPICE PROSPICE". The shield is surrounded by a laurel wreath.

KEYWORDS

Herbal medicine

Sutherlandia frutescens

Clinical trial

Type-2 diabetes

Protocol

Drug regulator

Ethics approval

Efficacy

UNIVERSITY of the
WESTERN CAPE

DECLARATION

I, declare that the thesis entitled: **The development of a study protocol, and ethics and regulatory approval documentation, for evaluation of the clinical efficacy of *Sutherlandia frutescens* in adult type-2 diabetics**, is my own work, has not been submitted before for any degree or examination in any other university, and that all the sources I have used or quoted have been indicated and acknowledged as complete references.

Ramadan Swead



Signed:

March 2018

UNIVERSITY of the
WESTERN CAPE

DEDICATION

I dedicate this master's thesis to my family, for their unending love, support and constant encouragement.



UNIVERSITY *of the*
WESTERN CAPE

ACKNOWLEDGEMENTS

In the name of Allah, the Most Beneficent, the Most Merciful

I make *shukr* to my Creator for granting me the inner strength and ability to fulfil my goals. I feel privileged to be chosen to make a contribution to the science world.

I would like to say *jazakallah* to my parents for all their support and understanding throughout my academic life. It is their love and constant encouragement that motivates me to do my best.

To Professor James A Syce, my supervisor, a big **THANK YOU** for believing in me and taking me under your wing. Your knowledge and love for what you do is inspiring. I am extremely grateful to you for providing me with the opportunity and honour to study under your guidance.

I would like to express my appreciation to Dr Halima Samsodien and Prof Hughes Gail for their support, advice and assistance to complete my work successfully.

Staff and colleagues at the School of Pharmacy and UWC for their presence and assistance at various stages of this research project.

My colleagues and friends at the School of Pharmacy and the Plant Medicine Research Group for their support and encouragement that made the completion of this thesis possible.

I am eternally grateful to you all. Thank you for your support and encouragement.

ABSTRACT

The prevalence of diabetes mellitus is increasing worldwide and it is becoming a significant medical problem in low- and middle-income countries. The condition can be controlled with a lifelong commitment to blood sugar monitoring, weight management, proper nutrition, exercise, and pharmacotherapy. Additional new pharmacotherapies are however needed to combat the increased prevalence and various traditionally used herbs, such as *Sutherlandia frutescens* (*S. frutescens*), are being advocated to supplement the management of type 2 diabetes mellitus. However, the clinical efficacy of *S. frutescens* in the management of type 2 diabetes mellitus has not yet been scientifically established.

The objectives of this study were to (1) establish a scientifically sound rationale for a clinical study to evaluate the efficacy of *S. frutescens* in adult type 2 diabetes mellitus, (2) design and develop a clinical trial protocol for a phase II efficacy study of *S. frutescens* in adult type 2 diabetics, and prepare (3) application documentation for the regulatory approval, and (4) prepare application documentation needed for ethics approval of an efficacy trial of *S. frutescens* in adult type 2 diabetics.

Firstly, literature on the traditional use and pharmacological and clinical studies of *S. frutescens*, herbal medicines used for diabetes mellitus and clinical trials of herbal medicines for diabetes mellitus were reviewed to establish a rationale and a design of a clinical study, to determine the efficacy of *S. frutescens* in type -2 diabetes mellitus. Next, the MCC's requirements for registration and regulatory control of clinical trials and, from a local South African Ethics Committee (Stellenbosch University), the requirements for proper ethical conduct of a trial was reviewed and used to compile a full protocol and ethics approval application (documentation) for a efficacy study of *S. frutescens* capsules in adult type -2 diabetics.

The results showed that *Sutherlandia frutescens* is a traditional herbal remedy commonly used in South Africa to treat diabetes, has anti-diabetic properties, Due to its versatility, it is a popular herb of choice for scientific scrutiny. *Sutherlandia frutescens* contains constituents, L-canavanine, -aminobutyric acid (GABA) and pinitol. Found in the leaf and seeds. Pinitol has been recognized for its anti-diabetic and immune modulating effects. *S. frutescens* has a long history of traditional use, with no reports of any serious side effects, suggesting that it can be considered as generally safe as shown in a study on vervet monkeys

(chlorocephalus aethiops) conducted by the South African Medical Research Council (MRC), it was reported that up to nine times the recommended dose of *Lessertia (L.) frutescens* (81 mg/kg body weight per day for three months) presented no significant changes in relevant haematologic, biochemical and physiological parameters. In conclusion, it was found that *S. frutescens* are non-toxic in vervet monkeys. Phase I of a safety clinical study on 25 healthy adults at Karl Bremer Hospital, Bellville, South Africa, showed that two 400 mg leaf powder capsules (800 mg per day) was well tolerated, with no side effects noted during or after the three months' trial period. There was no change in frequency of adverse events, or any clinically significant changes in most physical, vital, blood and biomarker indices. Therefore, the primary active compounds found in *S. frutescens* may possibly be acting synergistically in the human organism and mediating greater clinical benefit than has been noticed in the single compound studies. Based on the information on the traditional use, chemistry and pre-clinical studies of *S. frutescens*, and literature reported experiences on similar herbal products clinically tested in diabetics; a sound rationale for testing the efficacy of *S. frutescens* in adult type 2 diabetes patients was successfully established. Using information collected from clinical studies of conventional drugs (3 studies) and traditional herbal medicine (12 studies) in type-2 diabetes, and two *S. frutescens* studies (1 safety study in healthy volunteers and one HIV efficacy study), it was possible to develop a full protocol for testing the efficacy of *S. frutescens* in adult type 2 diabetes patients.

Collectively, using the information collected from reviews of traditional herbal medicine and *S. frutescens*, specifically, clinical trials done on healthy volunteer safety and HIV efficacy and completed clinical trials on herbal medicine (including 12 specific for type 2 diabetes and 3 clinical studies on conventional drugs specifically in type-2 diabetes). It was possible to develop a full protocol for testing the efficacy of *S. frutescens* in adult type 2 diabetes patients. A protocol for efficacy of *S. frutescens* was designed to do a 24- week randomized double-blind placebo-controlled study that could be conducted in South Africa. Moreover, the application for ethics committee and MCC committee was prepared for the protocol for approval for the afore-mentioned study.

The overall conclusion drawn from this study is that the study protocol was established according to the South African GCP guidelines and ready to be carried out to assess the efficacy of the traditional *S. frutescens* extract for the treatment of adult type 2 diabetes patients. Furthermore, the ethics and regulatory approval documentation can be used to

source funding (and/or collaboration) for the first study to determine/investigate the clinical efficacy of *S. frutescens* in adult type 2 diabetes patients.



UNIVERSITY *of the*
WESTERN CAPE

DEFINITIONS WITH REGARDS TO HERBAL MEDICINES AND CLINICAL TRIALS

For clarity and standardization, the following internationally accepted definitions are used for the below-mentioned terms in this thesis:

Herbal drugs are mainly whole, fragmented or cut plants or part of plants in an unprocessed state, usually in dried form, but sometimes fresh. Herbal drugs are precisely defined by the botanical scientific name according to the binomial system – genus, species, and variety. (World Health Organization. (2003)

African traditional medicine may be described as the total body of knowledge, techniques for the preparation and use of substances, measures and practices in use, whether explicable or not, that are based on the socio-cultural and religious bedrock of African communities, are founded on personal experience and observations handed down from generation to generation, either verbally or in writing and are used for the diagnosis, prevention or elimination of imbalances in physical, mental or social well-being (Akhtar R, 1987).

Traditional use of herbal medicine refers to the long historical use of these medicines.

A **clinical trial** is a research study in which a treatment or therapy is tested in people to see whether it is safe and effective (John Wiley & Sons, 2013). Each trial follows a **protocol** – i.e. a written, detailed plan that explains why there is a need for the study, what it is intended to do and how it will be conducted (Pocock, S. J. (2013). A **randomised trial** gives the best chance of knowing that the study results are caused by the treatment and not some other factor, such as people's choices or beliefs. Each participant in a randomised trial is assigned by chance (through a table of random numbers) to one of two groups:

- The investigational group, made up of people who will receive the therapy, also called the active treatment; or
- The control group, made up of people who will receive either the standard treatment (if there is one) for their disease or a placebo.

Trials can be **double blind**. This means that neither the researcher nor the participants know who has been assigned to which group. Blinding is another way to help minimize the chance of bias influencing the trial results.

A **placebo** is designed to resemble as much as possible the treatment being studied in a clinical trial, except that the placebo is inactive. (Wampold, B. E., Minami, et al, 2005).

A **patient diary** is a tool used during a clinical trial or a disease treatment to assess the patient's condition (e.g. symptom severity, quality of life) or to measure treatment compliance (Stone, Broderick, et al, 2004). Patient diaries are also used to find out if patients take the medication according to the treatment schedule, which is an important problem during clinical trials and the treatment of degenerative diseases with relatively few symptoms.



UNIVERSITY of the
WESTERN CAPE

Table of Contents

TITLE	i
KEYWORDS	ii
DECLARATION	iii
DEDICATION	iv
ACKNOWLEDGEMENTS	v
ABSTRACT	vi
DEFINITIONS WITH REGARDS TO HERBAL MEDICINES AND CLINICAL TRIALS	ix
LIST OF TABLES	xv
LIST OF FIGURES	xv
ABBREVIATIONS	xvi
Chapter 1: Introduction	1
1.1 The diabetes problem and the need for more treatment	1
1.2 Use of herbs for treating diabetes.....	2
1.2.1 <i>Sutherlandia</i> and its use in diabetes management	2
1.3 Clinical trials on herbal medicine in diabetes mellitus	3
1.3.1 Clinical efficacy trials on <i>Sutherlandia frutescens</i>	4
1.4 Control and regulation of clinical trials.....	4
1.4.1 Ethical requirements	5
1.4.2 Regulatory requirements.....	5
Chapter 2: Literature review	6
2.1 Introduction	6
2.2 Diabetes mellitus	6
2.2.1 Definition of diabetes mellitus	6
2.2.2 Types of diabetes mellitus	7
2.2.3 Pathophysiology	8
2.2.4 Complications	9
2.2.5 Diagnosis	9
2.2.6 Risk factors for type 2 diabetes mellitus	10
2.2.7 Treatment of type 2 diabetes mellitus.....	11
2.3 Challenges associated with type 2 diabetes therapy.....	13

2.4	Herbal medicine for diabetes mellitus.....	13
2.4.1	Herbs for treating type 2 diabetes mellitus.....	14
2.5	<i>Sutherlandia frutescens</i>	15
2.5.1	Botanical description.....	15
2.5.2	Chemical constituents of <i>S. frutescens</i>	16
2.5.3	Uses and pharmacological effects.....	19
2.5.4	Toxicology.....	20
2.6	Clinical trials.....	21
2.7	Regulatory control of herbal medicine.....	22
2.8	Ethics of herbal medicine.....	23
Chapter 3: Plan of Work.....		26
3.1	Study objectives.....	26
3.2	Hypotheses.....	26
3.3	Study approach.....	27
3.3.1	Determination of a scientific rationale to test <i>Sutherlandia frutescens</i> in type 2 diabetes mellitus.....	27
3.3.2	Design and development of a clinical trial protocol.....	27
3.3.3	Preparation of the application document for regulatory approval.....	28
3.3.4	Preparation of the application document for ethics approval.....	28
Chapter 4: A scientifically sound rationale for testing <i>Sutherlandia frutescens</i> in type 2 diabetes mellitus.....		30
4.1	Introduction.....	30
4.2	Methods.....	31
4.2.1	Review previous clinical studies of herbal medicine in the treatment of type 2 diabetes mellitus.....	31
4.2.2	The traditional uses and possible pharmacologically active compounds of <i>Sutherlandia frutescens</i>	31
4.2.3	Pre-clinical studies of <i>Sutherlandia frutescens</i> in diabetes mellitus.....	31
4.2.4	The safety data and dosage forms of <i>Sutherlandia frutescens</i>	32
4.3	Results and discussion.....	32
4.3.1	Review of previous clinical studies of herbal medicine in the treatment of type 2 diabetes mellitus.....	32
4.3.2	The traditional uses of <i>Sutherlandia frutescens</i> and its pharmacological active compounds.....	36
4.3.3	Pre-clinical studies of <i>Sutherlandia frutescens</i> in diabetes mellitus.....	37
4.3.4	Safety data and dosage forms of <i>Sutherlandia frutescens</i>	38
4.4	Conclusions.....	39

Chapter 5: Design and development of a clinical trial protocol to determine the efficacy of <i>Sutherlandia frutescens</i> in adult type 2 diabetes mellitus	40
5.1 Introduction	40
5.2 Methods.....	41
5.2.1 Review clinical trials of conventional anti-diabetic drugs for type-2 diabetes mellitus.....	41
5.2.2 Review of existing clinical studies of herbal products in type 2 diabetes mellitus	41
5.2.3 Preparation of a protocol to test the efficacy of <i>Sutherlandia frutescens</i> in adult type 2 diabetes mellitus.....	42
5.3 Result and discussion	42
5.3.1 Review of anti-diabetic drugs for type-2 diabetes mellitus.....	42
5.3.2 Review of existing clinical studies of herbal products in type 2 diabetes mellitus	45
5.3.3 A protocol for testing the efficacy of <i>Sutherlandia frutescens</i> in adult type 2 diabetes mellitus	48
5.4 Conclusion.....	52
Chapter 6: Application documentation required for regulatory approval of an efficacy trial of <i>Sutherlandia frutescens</i> in adult type 2 diabetes mellitus.....	53
6.1 Introduction	53
6.2 Methods.....	54
6.2.1 Determination of the general regulatory requirements for traditional herbal medicines	54
6.2.2 Preparation of an application for regulatory approval for testing <i>Sutherlandia frutescens</i> in type 2 diabetes mellitus	54
6.3 Results and discussion.....	54
6.3.1 The regulatory requirements for traditional herbal medicine.....	54
6.3.2 Application for regulatory approval for testing <i>Sutherlandia frutescens</i> in type 2 diabetes mellitus.....	58
6.4 Conclusions	59
Chapter 7: Application documentation for ethics approval for a clinical trial of <i>Sutherlandia frutescens</i> in type 2 diabetes mellitus.....	60
7.1 Introduction	60
7.2 Methods.....	60
7.2.1 Determination of the ethics requirements for an efficacy trial of <i>Sutherlandia frutescens</i> in type 2 diabetes mellitus to be conducted in South Africa	61
7.2.2 Preparation of an application for ethics approval to test <i>Sutherlandia frutescens</i> in type 2 diabetes mellitus.....	61
7.3 Results and discussion.....	62

7.3.1	Review of the ethics requirements an efficacy trial of <i>Sutherlandia frutescens</i> in type 2 diabetes mellitus	62
7.3.2	Application for ethics approval for testing efficacy of <i>Sutherlandia frutescens</i> in type 2 diabetes mellitus.....	64
Chapter 8: Conclusions and recommendations		65
References.....		67
APPENDIX 1: Complete Protocol.....		78
APPENDIX 2: Documentation for MCC – Clinical trial application.....		118
APPENDIX 3 A: Summary of requirements for ethics applications submitted to Stellenbosch university HREC		138
APPENDIX 3 B: Documentation for Stellenbosch Institutional Review Board – Application for registration of a research project		143



UNIVERSITY of the
WESTERN CAPE

LIST OF TABLES

Table 2. 1: Criteria for the diagnosis of diabetes mellitus	9
Table 4. 1: Clinical studies of herbal medicines for type 2 diabetes mellitus	33
Table 5. 1: Three clinical studies of anti-diabetic drugs for typ 2 diabetes mellitus	43
Table 5. 2: Clinical studies of herbal medicines for type 2 diabetes mellitus	46
Table 5. 3: Synopsis of protocol for testing the efficacy of <i>S. frutescens</i> on type 2 diabets melluits	49
Table 6. 1: A summary of regulatory requirements for the registration of traditional herbal medicines	55
Table 7. 1: Seven requirements for determining whether a research trial is ethical	63

LIST OF FIGURES

Figure 2. 1: <i>Sutherlandia frutescens</i> showing the fruit (a) and the flowers (b).....	16
--	----

ABBREVIATIONS

AIDS	Acquired Immune Deficiency Syndrome
CAM	Complementary and Alternative Medicine
CIOMS	Council for International Organizations of Medical Sciences
EBM	Evidence Based Medicine
EMA	European Medicines Agency
EU	European Union
FDA	Food Drug Administration
GABA	Gamma-Amino Butyric Acid
GCP	Good Clinical Practice
HIV	Human Immunodeficiency Virus
ICH	International Conference of Harmonization
IDDM	Insulin-Dependent Diabetes Mellitus
IP	Intellectual Property
IPT	Isoniazid Preventive Therapy
JAMA	Journal of the American Medical Association
MCC	Medicines Control Council
MRC	Medical Research Council
NCCAM	National Center for Complementary and Alternative Medicine
NIDDM	Non-insulin-Dependent Diabetes Mellitus
NIH	National Institutes of Health
NIH	National Institutes of Health
RCTs	Randomized Controlled Trials
REC	Research Ethics Committees
SAHSMI	South African Herbal Science and Medicine Institute
T2DM	Type 2 Diabetes Mellitus
THPs	Traditional Health Practitioners
ICIPS	International Center for Indigenous Phytotherapy Studies
TM	Traditional Medicine
WHO	World Health Organisation

Chapter 1

Introduction

1.1 The diabetes problem and the need for more treatment

Diabetes mellitus is a disease characterized by a combination of resistance to the action of insulin and insufficiency in insulin secretion (Deshpande, Harris-Hayes & Schootman, 2008) and is typically divided into two forms, viz. type 1 and type 2. It was estimated that 366 million people worldwide suffered from diabetes and that 80% of these diabetics lived in low- and middle-income countries (International Diabetes Federation, 2012). Generally, type 2 diabetes mellitus (T2DM) is the most common form of diabetes among adults, accounting for 90–95% of cases (Centers for Disease Control, 2011). In 2015 the estimated type 2 diabetes prevalence for South Africa was 7% for adults aged 20–79 years. (International Diabetes Federation, 2015).

Type 2 diabetes mellitus arises when the body produces insulin but does not use it effectively, so that glucose builds up in the bloodstream, instead of being absorbed by the cells (National Institutes of Health (NIH), 2014). The aforementioned phenomenon is also known as insulin resistance. In type 2 diabetes mellitus hyperglycaemia is a major barrier to achieving treatment goals and has been shown to be linked to an increased risk of cardiovascular events, dementia, and can contribute to weight gain. T2DM is also associated with serious long-term microvascular and macrovascular complications such as cardiovascular disease, neuropathy and diseases of the eyes, which, in turn, increase morbidity, mortality and healthcare costs (Cade, 2008). The general treatment of T2DM includes healthy eating, regular exercise, diabetes medication or insulin therapy and blood-sugar monitoring (Marić, 2010).

Presently, there is no cure for T2DM, but the condition can be partially controlled with lifelong commitment to blood-sugar monitoring, weight management, proper nutrition and physical exercise, all making diabetes a very restrictive condition. Moreover, diabetes mellitus tends to progress with age and medications are also often needed. These medications typically include oral hypoglycaemics, non-insulin injectable hypoglycaemic medication and insulin (Evans &

Rushakoff 2016). For instance, the oral hypoglycaemic metformin is generally used in patients with T2DM and is considered as the “gold standard” (Marić, 2010). However, these drugs (and other anti-diabetic medicine) have significant adverse effects and there is a need to explore other alternatives for treatment of this condition. Such alternatives may include herbal medication such as *Sutherlandia frutescens*.

1.2 Use of herbs for treating diabetes

According to Ghorbani (2013), many herbs have been recommended for treating diabetes, but in most cases these recommendations are only based on results of animal studies, and limited evidence exist about their clinical usefulness. However, based on the clinical studies identified in a systematic review conducted by Ghorbani (2013), the plants, *Aegle marmelos*, *Allium cepa*, *Gymnema sylvestre*, *Momordica charantia*, *Ocimum sanctum*, *Nigella sativa*, *Ocimum sanctum*, *Panax quinquefolius*, *Salacia reticulate*, *Silybum marianum* and *Trigonella foenum-graecum* were shown to display hypoglycaemic and, in some cases, hypolipidaemic activities in diabetic patients. In addition to the above, the South African medicinal plant, *Sutherlandia frutescens* (*S. frutescens*), has also been claimed to have anti-diabetic properties (Williams, et al 2013 and Ojewole (2004) and Chadwick et al. (2007), but it was not listed in the systematic review of Ghorbani (2013) and this activity has not yet been clinically proven.

1.2.1 *Sutherlandia* and its use in diabetes management

According to van Wyk and Albrecht (2008), *S. frutescens* (tribe Galegeae, Fabaceae), is a popular plant used in traditional medicine (TM) and is indigenous to South Africa, Lesotho, Southern Namibia and South-Eastern Botswana. There are many traditional uses for *S. frutescens*, including that of treatment of poor appetite, indigestion, stomach complaints, dysentery, colds, influenza, kidney conditions, fever, diabetes, internal cancers, uterine troubles, liver conditions, backache, rheumatoid arthritis, urinary tract infections, stress, anxiety, dropsy and heart failure (Van Wyk & Albrecht, 2008).

One of the traditional uses of *S. frutescens* is in the treatment of type-2 diabetes and a few pre-clinical studies have also been conducted to provide further scientific support for these traditional uses. For instance, (Williams, et al 2013) investigated the capacity of an aqueous extract of *S. frutescens* in the prevention of insulin resistance in a human liver cell culture and

its effect on diabetes-related genes. Their study found that *S. frutescens* could prevent insulin resistance, a precursor of T2DM, in hepatocytes. In addition, Ojewole (2004) presented evidence that *Sutherlandia* extracts can reduce glucose uptake in streptozotocin (STZ) - treated mice while Chadwick et al. (2007) showed statistically significant increases in glucose uptake and no weight gain in pre-diabetic rats receiving *Sutherlandia* via their drinking water. *S. frutescens* also revealed various bioactive compounds previously identified in its aqueous extracts. For instance, it is also known that *S. frutescens* contains pinitol, a known anti-diabetic agent, which may have an application in treating diabetes (Ostlund & Sherman, 1996). Therefore there is some pre-clinical study evidence suggesting that *Sutherlandia* extracts showed promise as a medication for T2DM, but the clinical efficacy and mechanism(s) of action of this herbal medicine need further study (Chadwick, et al. 2007) and for this a clinical study is required.

1.3 Clinical trials on herbal medicine in diabetes mellitus

A clinical trial is any systemic study of a medicinal product in human subjects. It can be conducted in patients or in non-patient (i.e. healthy) volunteers to determine the absorption, distribution, metabolism and excretion of investigational products and to ascertain its efficacy and safety or identify any adverse reaction (Black, 1996).

There are few clinical trials of good quality that have reported the efficacy of herbal medicine in treating various conditions in human subjects, but clinical trials on the effect of herbal medicine in the glycaemic control of T2DM have been reported. For instance, in a systematic review reported by Suksomboon, et al. (2011), several clinical trials using various herbal remedies, which included *Cinnamomum cassia* (cinnamon), *Ipomoea batatas* (sweet potato), *Silybum marianum* (milk thistle) and *Trigonella foenum-graecum* (fenugreek), were mentioned. Moreover, it was found that the use of sweet potato, milk thistle and fenugreek, for at least eight weeks, may improve glycaemic control and it was concluded that supplementation with these herbs, in addition to dietary control or medications, may offer an alternative for T2DM patients who cannot achieve their target glycaemic control. The quality of some of the trials on herbal remedies is of high quality but many trials of herbal treatments have exhibited poor quality, due to lacking an intention to treat analysis or a comment on whether the trial

protocol was successful (Sharma, & Gupta, R. 2010). Overall however, sufficient traditional use and pre-clinical data on several herbal medicines are now available to justify the conduct of properly controlled clinical studies of these medicines, especially with respect to their possible use in diabetes.

1.3.1 Clinical efficacy trials on *Sutherlandia frutescens*

A review of the literature revealed that no clinical trial on the efficacy of *S. frutescens* in diabetes mellitus has thus far been conducted, but that such a trial is warranted. Indeed, sufficient traditional and pre-clinical data on this traditional medicine is now available to justify the conduct of a controlled clinical study of this herb in diabetes. In addition, results of animal studies (Seier, et al., 2002), an MRC study in monkeys as well as a Phase I safety clinical study (Johnson, & Folk, 2007) and a Phase II efficacy study (Wilson *et al.*, 2015) of *Sutherlandia* in HIV infected patients have shown no indications of significant toxicity.

The first step for conducting a clinical efficacy study of *Sutherlandia* in diabetes mellitus is, however, to develop a protocol. The latter is vital as it describes the collection of activities involved in performing the trial and acts as a point of referral. Firstly, it specifies the treatments that will be studied and the process and method of their usage and administration. In addition, the background, rationale, objectives and design of the study, the methodology to be employed, the data analysis and statistical considerations as well as the overall organization of the clinical study is described in a protocol. And, such a protocol is needed to determine the clinical efficacy of *S. frutescens* in T2DM.

1.4 Control and regulation of clinical trials

Worldwide, the conduct of clinical trials is a highly regulated and tightly controlled activity. The same applies in South Africa, i.e. also here the control and regulation of clinical trials is managed by certified human subjects ethics committees, and the drug regulatory authority, viz. the Medicines Control Council (MCC) in the case of South Africa. An ethics committee has to approve the trial (i.e. certify that it is conducted in accordance with current acceptable ethical rules or norms) while the regulatory body has to provide permission for the trial to be conducted on the (experimental) medicinal product. Until these processes have been completed, a trial cannot begin with subject recruitment or any other steps involved in conduct of the trial.

1.4.1 Ethical requirements

According to the Guidelines for Good Practice (GGP) in the Conduct of Clinical Trials with Human Participants in South Africa (Department of Health, 2006), “all clinical trials conducted in South Africa must undergo ethical review by an accredited ethics committee.” An ethical review affects potential participants and the general day-to-day functioning of the health system. Therefore, the main responsibility of a Research Ethics Committee (REC) in South Africa is to ensure the protection of participants, the respect of their rights and their safety and well-being when involved in a trial. Along with providing public assurance of this protection via reviewing, approving and providing comment on clinical trial protocols, the suitability of investigator(s), facilities, and methods and procedures which are used to obtain informed consent (Department of Health, 2006), the Research Ethics Committee (REC) is itself also required to be accredited by the National Health Research Ethics Council (NHREC).

1.4.2 Regulatory requirements

Generally, Section 5.2 of the International Conference on Harmonisation (ICH), and Good Clinical Practice (GCP) guidelines, E6 (detail of the ref), stipulates that before initiating a clinical trial, the sponsor (or the sponsor and the investigator, if required by the applicable regulatory requirement) should submit any required application to the appropriate authority for review, acceptance, and/or permission (as required by the applicable regulatory body) (Department of Health, 2006). In South Africa, the regulatory body is the MCC and the South African GCP guidelines state that all clinical trials of both non-registered medicinal substances (such the herbal medicine *S. frutescens* in this study) and for new indications of registered medicinal substances must be reviewed by the MCC.

Given the above arguments, the objectives of this study consequently were to, firstly, establish a scientifically sound rationale for testing the efficacy of *S. frutescens* in T2DM, then design and develop a clinical trial protocol for testing the efficacy of *S. frutescens* in adult T2DM and, finally, prepare the required application documentation for regulatory and ethics approvals of such a trial.

Chapter 2

Literature review

2.1 Introduction

In this chapter, brief review of the definition of diabetes and types of diabetes, the pathophysiology, diagnosis, current treatment and challenges of type 2 diabetes mellitus (T2DM) therapy as well as the selected plant species *Sutherlandia frutescens* (*S. frutescens*) are given. In addition, an overview of clinical trials and regulation and ethical considerations of herbal medicine are presented.

2.2 Diabetes mellitus

2.2.1 Definition of diabetes mellitus

Diabetes mellitus, or simply diabetes, is a group of diseases characterized by high blood glucose levels that result from defects in the body's ability to produce and/or use insulin. It is a condition primarily defined by the level of hyperglycaemia giving rise to a risk of microvascular damage (retinopathy, nephropathy and neuropathy). Diabetes mellitus may present with characteristic symptoms such as thirst, polyuria, blurring of vision and weight loss. Often symptoms are not severe or may be absent. Furthermore, it is associated with reduced life expectancy, significant morbidity attributed to specific diabetes-related microvascular complications, increased risk of macrovascular complications (ischaemic heart disease, stroke, and peripheral vascular disease) and diminished quality of life (IDF, 2015).

Diabetes mellitus continues to pose a substantial global healthcare challenge with marked medical and socioeconomic ramifications. For instance, recent estimates from the International Diabetes Federation (IDF, 2015) suggests that 415 million adults, worldwide, were living with diabetes in 2015 (1 in 11 adults) and that as many as 642 million people (1 in 10 adults) may be affected by the year 2040 (IDF, 2015) (<http://www.diabetesatlas.org>).

2.2.2 Types of diabetes mellitus

The first widely accepted classification of diabetes mellitus was published by the World Health Organization (WHO) in 1980 (WHO, 1999). Two major classes of diabetes mellitus were proposed, viz. insulin-dependent diabetes mellitus (IDDM or Type 1) and non-insulin-dependent diabetes mellitus (NIDDM or Type 2). Other types, as well as gestational diabetes, were also included. The modified version of the 1985 report (Diabetes Mellitus: WHO, (1985) Report of a Study Group, was widely accepted and is used internationally. The recommendations that the terms, IDDM and NIDDM, as classification according to treatment should no longer be used and that the terms ‘Type 1’ and ‘Type 2’ rather be used to describe the pathogenesis of diabetes were then accepted later on (Ritov *et al.*, 2010 and WHO, 1999).

2.2.2.1 Type 1 diabetes mellitus

Type 1 diabetes mellitus (T1DM) indicates the process of beta-cell destruction in the pancreas that may ultimately lead to diabetes mellitus in which “insulin is required for survival” to prevent the development of ketoacidosis, coma and death. T1DM account for only about 5–10% of all cases of diabetes; however, its incidence continues to increase worldwide and this has serious short-term and long-term implications (WHO, 1999). Management of T1DM diabetes is best undertaken in the context of a multidisciplinary health team and requires continuing attention to many aspects, including insulin administration, blood glucose monitoring, meal planning and screening for diabetes-related complications. These latter complications mainly consist of microvascular and macrovascular diseases, which account for the major morbidity and mortality complications associated with T1DM (American Diabetes Association, 2009; Daneman, 2006).

2.2.2.2 Type 2 diabetes mellitus

Type 2 diabetes mellitus (T2DM) is a serious global health issue resulting from defects in insulin secretion. The WHO estimates that the prevalence of diabetes worldwide would reach 366 million in 2030 (Fakhoury *et al.* 2010; Rodbard *et al.* 2007). Generally Type 2 diabetes accounts for approximately 90–95% of all cases (Rodbard *et al.*, 2007) and 80% of these diabetics live in low- and middle-income countries (IDF, 2012). Approximately 6.6% of the world’s population aged between 20 and 79 years has T2DM and this estimation is projected to increase to 7.8% by 2030 (Atlas, 2006; Fakhoury *et al.*, 2010). T2DM is associated with serious long-term macrovascular complications such as cardiovascular disease, neuropathy and diseases of the eyes, which in turn, increase morbidity, mortality and healthcare costs (Cade,

2008). There is no cure for diabetes, but the condition can be controlled with a lifelong commitment to blood-sugar monitoring, weight management, nutrition and exercise making it very restrictive. General treatment of T2DM includes healthy eating, regular exercise, diabetes medication or insulin therapy and blood-sugar monitoring.

People with diabetes are at a greater risk of developing cardiovascular diseases such as heart attack and stroke if the disease is left undiagnosed or poorly controlled. They also have elevated risks for sight loss, foot and leg amputation caused by damage to the nerves and blood vessels and renal failure requiring dialysis or transplantation (Lawrence, Conrad & Moore, 2012). Before people develop T2DM, they almost always have ‘prediabetes’, i.e. blood-glucose levels that are higher than normal but not yet high enough to be diagnosed with diabetes. Recent research has shown that some long-term damage to the body, especially the heart and circulatory system, may already be occurring during prediabetes (American Diabetes Association, 2009 and DePaula *et al.*, 2008).

2.2.3 Pathophysiology

An understanding of the pathophysiology of diabetes rests upon knowledge of the basics of carbohydrate metabolism and insulin action (Ozougwu, 2013). Following the consumption of food, carbohydrates are broken down into glucose molecules in the gut. Glucose is absorbed into the bloodstream elevating blood glucose levels. This rise in glycaemia stimulates the secretion of insulin from the beta cells of the pancreas. Insulin is needed by most cells to allow glucose entry. Insulin binds to specific cellular receptors and facilitates entry of glucose into the cell, which uses the glucose for energy. The increased insulin secretion from the pancreas and the subsequent cellular utilization of glucose results in lowering of blood glucose levels. Lower glucose levels then result in decreased insulin secretion. If insulin production and secretion are altered by disease, blood glucose dynamics will also change. If insulin production is decreased, glucose entry into cells will be inhibited, resulting in hyperglycaemia. The same effect will be seen if insulin is secreted from the pancreas but is not used properly by target cells. If insulin secretion is increased, blood glucose levels may become very low (hypoglycaemia) as large amounts of glucose enter tissue cells and little remains in the bloodstream. Multiple hormones may affect glycaemia. Insulin is the only hormone that lowers blood glucose levels, but the counter-regulatory hormones such as glucagon, catecholamines,

growth hormone, thyroid hormone, and glucocorticoid all act to increase blood glucose levels, in addition to their other effects (Ozougwu, 2013 & Scheen, 2003).

2.2.4 Complications

Complications caused by diabetes are a major cause of disability, reduced quality of life, and death. Diabetes complications can affect various parts of the body manifesting in different ways for different people. In men, these are responsible for erectile dysfunction, low testosterone levels and emotional factors, such as depression, anxiety or stress. In women, they are accountable for depression and frequent eating disorders. Even in women who do not have diabetes, pregnancy brings about the risk of gestational diabetes. In addition, diabetes can affect every part of the body, including the feet, the eyes, and the skin. In fact, such problems are sometimes the first signs that a person has diabetes. Foot complications can get worse and lead to serious complications, such as neuropathy, skin changes, calluses as well as foot ulcers and poor circulation (Loomans *et al.*, 2004),(Vaz & Patnaik, 2012)(Aalto, Uutela & Aro, 1997).

2.2.5 Diagnosis

The diagnosis of diabetes mellitus is easily established when a patient presents the classic symptoms of hyperglycaemia and has a fasting plasma glucose ≥ 7.0 mmol/l or 2-hour plasma glucose ≥ 11.1 mmol/l (WHO, 1999). The criteria for the diagnosis of diabetes are shown in Table 2.1.

Table 2. 1: Criteria for the diagnosis of diabetes mellitus

Criterion 1		Criterion 2		Criterion 3
Symptoms of diabetes and casual plasma glucose ≥ 200 mg/dl (11.1 mmol/l).	Or	Fasting Blood Glucose(FPG) ≥ 126 mg/dl (7.0 mmol/l).	Or	2-hour plasma glucose ≥ 200 mg/dl (11.1 mmol/l) during an oral glucose tolerance test (OGTT).
Casual is defined as any time of day without regard to time since last meal. The classic symptoms of diabetes include polyuria, polydipsia, and unexplained weight loss.		Fasting is defined as no calorie intake for at least 8 hours.		The test should be performed as described by the World Health Organization (WHO), using a glucose load containing the equivalent of 75 g anhydrous glucose dissolved in water.

The following tests are done for the basic diagnosis of T2DM. (1) The **fasting plasma glucose (FPG)** test which measures blood glucose in a person who has not eaten anything for at least 8 hours. This test is used to detect diabetes and prediabetes. (2) **An oral glucose tolerance test (OGTT)** which measures blood glucose after a person fasts for at least 8 hours, the zero hour reading, and two hours later, after the person has drunk a 75 g glucose-containing beverage. This test can be used to diagnose diabetes and prediabetes. Generally, the FPG test is the preferred test for diagnosing diabetes because of its convenience and low cost. However, this test may miss diabetes or prediabetes cases which can be determined with the OGTT. The FPG test is most reliable when done in the morning. Research has shown that the OGTT is more sensitive than the FPG test for diagnosing prediabetes, but less convenient to administer. (3) The **random plasma glucose test**, also called a **casual plasma glucose test**, measures blood glucose without regard to when the person being tested last ate. This test, along with an assessment of symptoms, is used to diagnose diabetes but not prediabetes. Test results indicating that a person has diabetes should be confirmed with a second test on a different day (American Diabetes Association, 2009; WHO, 1999).

2.2.6 Risk factors for type 2 diabetes mellitus

There are controllable risk factors associated with T2DM, including obesity, diet and an inactive lifestyle. However, other uncontrollable risk factors, such as genetics, increasing age, insulin resistance, ethnicity and family history of diabetes also play a dramatic role. As concerns genetic factors, research has shown that certain gene variations raise the risk of developing diabetes. These genes can be associated with insulin sensitivity in the body's tissues, decreased insulin production and an increased risk of obesity. Race and ethnicity, on the other hand, are responsible for higher levels of diabetes in certain ethnic groups. This is partly because of higher rates of high blood pressure, obesity and diabetes in these populations (Boulton *et al.*, 2005).

Although genes and ethnicity are risk factors for T2DM, they are not the major determinants of whether someone develops the disease. Changes in diet and decreased physical activity related to rapid technological development and urbanisation have led to a sharp increase in the number of people developing diabetes. A history of substance use has been reported as a significant factor associated with earlier age of onset of T2DM. Illicit drug use has also been associated with diabetes, according to research in the United States (Karlson, Mohsen &

Cherpitel, 2001). The most common drugs used regularly, are marijuana and cocaine, while other illicit drugs (amphetamines, heroin, hallucinogens, and nonmedical inhalants) are often used by diabetic primary care patients.

2.2.7 Treatment of type 2 diabetes mellitus

2.2.7.1 Insulin therapy

Insulin is a hormone that treats diabetes by controlling the amount of sugar (glucose) in the blood. When used as a medication, it is derived from either pork (porcine), beef or is genetically made to be identical to human insulin (Buysschaert, Dramaix, Wallemacq & Hermans, 2000). Patients with T1DM depend on external insulin for their survival, because the hormone is no longer produced internally. Certain patients with T2DM may eventually require insulin if other medications fail to control blood glucose levels adequately.

There are many types of insulin used to treat diabetes. They are classified by how fast they start to work, when they reach their “peak” level of action (i.e. when the concentration of insulin in the blood is highest) and how long their effects last.

The types of insulin include:

- **Rapid-acting insulin**, which starts working within a few minutes and lasts for a couple of hours.
- **Regular- or short-acting insulin**, which takes about 30 minutes to work and lasts for 3 to 6 hours.
- **Intermediate-acting insulin**, which takes 2 to 4 hours to work and its effects can last for up to 18 hours.
- **Long-acting insulin**, which takes 6 to 10 hours to reach the bloodstream, but can keep working for an entire day (Tuomilehto, 2001).

Insulin for diabetes can be injected under the skin (subcutaneously) or into the vein (intravenously). Subcutaneous insulin injections continue to be the mainstay of therapy for all people with T1DM and individuals with T2DM. Insulin can be injected using a needle and syringe, a cartridge system or a prefilled pen system or an insulin pump may be used.

The initial dose is calculated based on the patient’s weight and sensitivity to insulin, which varies from person to person. When given under the skin, insulin is typically taken so that two-thirds of the total daily dose is given in the morning and one-third of this given in the evening (Skyler, Skyler, Seigler, & O’Sullivan, 1981).

The major side effects of insulin in treating diabetes include low blood sugar (hypoglycaemia), hypertrophy (i.e. enlargement of the area of the body that has received too many insulin injections) and a rash at the site of injection or over the entire body. Other known side effects are rare. (Gkaliagkousi, 2007).

2.2.7.2 Oral hypoglycaemic agents

The term, oral hypoglycaemic agent, can refer to any anti-diabetic medication. The following five categories will be presented in brief: sulphonylureas and similar (secretagogues), biguanides (sensitizers), thiazolidinediones, alpha-glucosidase inhibitors, and incretineanalogues/agonists (Boulton *et al.*, 2005).

The secretagogues like sulphonylureas block the adenosine triphosphate (ATP)-sensitive K^+ channel in the beta cells of the pancreas and as a result, they stimulate insulin secretion. They are usually indicated as the first choice once a patient is diagnosed with T2DM. Their side effects include hypoglycaemia, a disulfiram reaction and possible drug interactions resulting from the competition for protein binding/metabolism/secretion. The meglitinides are short-acting secretagogues, similar to the sulphonylureas. They also block the ATP-sensitive K^+ channel and they open the Ca^{2+} channels, thus stimulating insulin secretion. Their side effects include weight gain and hypoglycaemia. Biguanides reduce the hepatic synthesis and the output of glucose, while they increase the insulin uptake in the skeletal muscle. They are actually anti-hyperglycaemic and not hypoglycaemic and are indicated for T2DM and polycystic ovary syndrome. They also help reduce low-density lipoprotein (LDL) cholesterol and triglyceride levels and may help with weight loss. Their side effects include *lactate* acidosis, GIT discomfort, diarrhoea, renal toxicity, and they do not cause hypoglycaemia. Thiazolidinediones are selective agonists of peroxisome proliferator-activated receptors ($PPAR\gamma$), thus activating the insulin-sensitive genes regulating glucose and fat metabolism. As a result, they increase the insulin sensitivity in the peripheral tissue. Their main side effect include hepatotoxicity. Inhibitors of the alpha-glucosidase or the alpha amylase reduce the intestinal absorption of starch, dextrans, disaccharides and as a result, they reduce the postprandial plasma glucose. They are indicated for T1DM and T2DM in combination with diet and insulin. Their side effects include malabsorption, flatulence, and diarrhoea. Peptide analogues such as the glucagon-like peptide-1 (GLP-1) agonists bind to a membrane GLP-1 receptor. They are metabolized by the dipeptidyl peptidase-IV enzyme (DPP-IV) and their side effects include nausea, hypoglycaemia (if given together with insulin secretagogue) and

exenatide–acute pancreatitis. The mode of action of the dipeptidyl peptidase-4 (DPP-4) inhibitors (gliptines) is that they increase the blood concentration of the incretin GLP-1 (glucagon-like peptide-1) by inhibiting its degradation by dipeptidyl peptidase-4 (DPP-4). Consequently, they produce fewer side effects than the other oral hypoglycemic agents (OHA), but the fact that they constitute a new class of drugs should be taken into account (Gkaliagkousi, 2007).

2.3 Challenges associated with type 2 diabetes therapy

The current methods of treatment for T2DM do not address the causes of the disease and have side effects. Approaches to the treatment of this chronic progressive disease include diet modification and oral hypoglycaemic medications. These approaches have generally proven to be inadequate, as baseline glycosylated haemoglobin remains relatively high in subjects diagnosed with diabetes and on treatment, while insulin therapy only solves the problem temporarily (García-Pérez, Álvarez, Dilla, Gil-Guillén & Orozco-Beltrán, 2013). Even with the latest pharmaco-therapies, patients continue to develop macro- and microvascular complications. Thus, there is an obvious need for suitable alternative treatment methods, effective treatment options and improved treatment guidelines. The search continues for an ideal anti-diabetic drug that will not only normalize blood glucose, but also provide beta-cell rest and possibly restoration of its function (Vaz & Patnaik, 2012).

2.4 Herbal medicine for diabetes mellitus

The WHO has recognized the important contribution of Traditional Medicine (TM) to provide essential care (World Health Organization, http://www.who.int/topics/traditional_medicine/en/ last access: May 2016). In 1989, the U.S. Congress established the Office of Alternative Medicine within the National Institutes of Health to encourage scientific research in the field of TM (<http://nccam.nih.gov>, last access: May 25, 2016). The European Scientific Cooperative on Phytotherapy was founded in 1989 with the aim of advancing the scientific status and harmonization of phytomedicines throughout Europe (www.escop.com, last access: May 25, 2016). These led to an increase in investment in the evaluation of herbal medicines. In the United States, the National Center for Complementary and Alternative Medicine at the National Institutes of Health spent approximately US\$33 million on herbal medicines in the fiscal year 2005. In 2004, the National Canadian Institute committed nearly US\$89 million on studying a range of traditional therapies. The scale of investment is low in comparison to the

total research and development expenses of the pharmaceutical industry. This is mainly because, unlike synthetic chemicals, herbs cannot be patented. Therefore, there is little money to be made by funding such research. Nevertheless, this reflects genuine public, industry and governmental interest in this area (Li & Vederas 2009).

Several important factors have contributed to the growth of this worldwide phytotherapeutic market. Among these are the following: consumer preference for natural therapies; concern regarding undesirable side effects of modern medicines and the belief that herbal drugs are free of side effects; a great interest in alternative medicines; preference of populations for preventive medicine attributable to increasing population age; the belief that herbal medicines might be more effective and beneficial in treating certain diseases where conventional therapies and medicines have proven to be inadequate; and the high cost of synthetic medicines. (Pan *et al.*, 2013)

That more than 80% of drug substances are either directly derived from natural products or developed from a natural compound has been widely accepted (Dias *et al.*, 2012). During 1981 to 2006, 47.1% of a total of 155 clinically approved anti-cancer drugs were derived from natural products in North America, Europe and Japan (Newman & Cragg, 2007). In China, at least 130–140 new drugs, either single chemical entities extracted from herbal medicines, or synthetically modified compounds are currently in clinical use (Dias *et al.*, 2012). In general, herbal medicines with a long history and theory of use in clinical settings should be more promising candidates for drug discovery (Pan *et al.*, 2013).

2.4.1 Herbs for treating type 2 diabetes mellitus

According to Ghorbani (2013), many herbs have been recommended for treating diabetes but, in most cases, the recommendations are based on animal studies and limited evidence exist about their clinical usefulness. Various herbs have been used to manage diabetes. Based on the clinical studies identified in a systematic review conducted by Ghorbani, (2013) *Aegle marmelos*, *Allium cepa*, *Gymnema sylvestre*, *Momordica charantia*, *Ocimum sanctum*, *Nigella sativa*, *Ocimum sanctum*, *Panax quinquefolius*, *Salacia reticulate*, *Silybum marianum* and *Trigonella foenum-graecum* displayed hypoglycemic and, in some cases, hypolipidemic activities in diabetic patients. In addition to the above, the South African medicinal plant, *S. frutescens*, was also claimed to have anti-diabetic properties (Ojewole, 2008), but it was not listed in the systematic review by Ghorbani (2013).

2.5 *Sutherlandia frutescens*

Sutherlandia frutescens is one of the most commonly used traditional herbal remedies in South Africa (Fernandes et al., 2004; Ojewole, 2008). Widely known as an anti-diabetic and cancer bush, this versatile medicinal herb is indigenous to Africa (Ojewole, 2008). It is found in the southern parts of Namibia, the extreme south-eastern corner of Botswana, the western, central and eastern parts of South Africa and most of Lesotho (Williams, et al 2013 and Ojewole (2004)). Because of the wide distribution range of *S. frutescens*, it is known by several vernacular names in different languages. In English, it is known as *cancer bush*, in Afrikaans, as *kankerbos*, in Sesotho the plant is known as *musa-pelo* and *motlepelo*, while in Setswana it goes by the name, *phetola*. In isiZulu, *insiswa*, and in isiXhosa, *unwele*, are the names which refer to *S. frutescens* (van Wyk & Albrecht, 2008).

Sutherlandia frutescens belongs to:

- Division : Magnoliophyta
- Class : Magnoliopsida
- Order : Fabales
- Family : Fabaceae
- Subfamily : Faboideae
- Tribe : Galegeae
- Genus : *Sutherlandia*
- Species : *S. frutescens*

(Van Wyk & Gericke, 2000)

2.5.1 Botanical description

Sutherlandia frutescens is a perennial flowering shrub which grows up to 2 m in height (van Wyk & Wink, 2004). The stems are prostrate to erect with compound pinnate leaves and leaflets that are 4–10 mm long, grey-green in colour and are oblong to linear elliptic. The leaves are three or more times longer than wide, slightly densely hairy and silvery in appearance and have a very bitter taste. The orange-red flowers, up to 35 mm long, are carried in short racemes in the leaf axils at the tips of the branches and flower in spring to mid-summer (September–

December). The wing petals of these flowers are very small and concealed in the calyx, with the standard petal being much shorter than the keel, and thus not the typical 'pea' flower (Gericke *et al.*, 2001; van Wyk & Albrecht, 2008). The seeds are black, flattened and approximately 3 mm in diameter (Fig 2.1) (Van Wyk, & Albrecht, 2008). The aerial parts of the plant, most commonly the leaves and twigs, are utilized in the treatment of various ailments.



Figure 2. 1: *Sutherlandia frutescens* showing the fruit (a) and the flowers (b)

<http://kumbulanursery.co.za/plants/lessertia-frutescens-previously-sutherlandia-frutescens>, access date 17Jul 2017)

2.5.2 Chemical constituents of *S. frutescens*

Various chemical constituents have been identified in *S. frutescens*, which contribute to the efficacy of this medicinal plant including the non-protein amino acid L-canavanine, *gamma*-aminobutyric acid (GABA) and flavonoids, pinitol and asparagine. In addition a triterpenoid glucoside and have been isolated and characterized (van Wyk & Albrecht, 2008).

2.5.2.1 Free amino acids

The leaves of *S. frutescens* contain high levels of free and protein bound amino acids (Moshe, 1998; van Wyk, 2008). Analysis of the leaves of commercial *S. frutescens* grown at different localities show high levels of the free amino acids; asparagine (1.6-35.0 mg/g), proline (0.7-7.5 mg/g) and arginine (0.5-6.7 mg/g) (van Wyk and Albrecht, 2008). The presence of arginine is of particular importance because of its biological activities.

Arginine is considered a non-essential amino acid since the body produces it naturally (Boom *et al.*, 2008). Free arginine is an important signalling molecule in both animals and plants. In

plants, arginine is converted to nitrous oxide (NO) by a group of enzymes called nitric oxide synthases. NO can also be synthesized from nitrite using nitrate reductase (Bethke, Bodger, & Jones, 2004). NO is an important signalling molecule that acts on many tissues to regulate a wide range of physiological and cellular processes. The endogenous production of NO plays a vital role in influencing physiological processes (Abramson, 2005) such as vasodilation. NO production is suggested to be beneficial in the treatment or management of illness conditions that improve with vasodilation (Boom *et al.*, 2008). Increasing evidence indicates that NO also plays a complex role in modulating the inflammatory response (Abramson, 2005). Arginine is also active against viral infections, including *Coxsackie* virus because of its contribution to NO synthesis. However, increased NO production can lead to NO-induced cytotoxicity by oxidative injury, resulting in cellular and organ dysfunctions. Thus, arginine may be beneficial or harmful to the body and the question of its safety could be related to the dose administered (Sia, 2004).

2.5.2.2 L-canavanine and GABA

2.5.2.2.1 L-canavanine

L-canavanine is a common compound recorded in members of the legume family (van Wyk & Albrecht, 2008) and is one of three principle constituents of *S. frutescens* (Mills *et al.*, 2005). High levels of L-canavanine in *S. frutescens* leaves were discovered by Moshe (1998). The levels of L-canavanine in the leaves vary from 0.42 mg/g to 14.5 mg/g (Moshe, 1998) and about 1.3-3.1 mg/g dry weight in commercial preparations of *S. frutescens* (van Wyk, 2008). L-canavanine is a compound that may be responsible for the anti-cancer and anti-inflammatory properties ascribed to *S. frutescens*. L-canavanine also has an anti-cancer capacity that has been found to abate pancreatic cancer (Sia, 2004). Its major metabolite, canaline is being developed as a potentially new anti-cancer drug for pancreatic cancer (Reid *et al.*, 2006). L-canavanine is reported to have anti-viral activities against influenza and retroviruses including HIV (Mills *et al.*, 2005).

2.5.2.2.2 GABA

GABA is both an amino acid and an inhibitory neurotransmitter that acts outside the central nervous system (Sia, 2004; Mills *et al.*, 2005). It is a non-protein amino acid that is biologically important (Chemistry 108B, 2005) as it is responsible for most of the brain's fast inhibitory transmission (Jocham & Ullsperger, 2008). According to van Wyk and Albrecht (2008) commercial samples of *S. frutescens* contain 0.23-0.85 mg/g of GABA. The presence of GABA

could partly account for *S. frutescens*' use in the treatment of stress and anxiety (van Wyk & Albrecht, 2008). Stress-related ailments are known to be linked to the endocrine system. Tai *et al.* (2004) suggested that GABA could play a role in elevating the mood of patients suffering from a chronic illness. Prevoo *et al.* (2008) recently showed that *S. frutescens* may reduce the adrenal P450 enzyme, and thus indicated a possible mechanism by which symptoms of stress and glucocorticoid levels can be reduced.

2.5.2.3 Flavonoids

In an attempt to provide chemical markers for the aerial parts of *S. frutescens*, a phytochemical investigation done by Avula *et al.* (2010) led to the identification of four flavonoids, *Sutherlandin A*, *Sutherlandin B*, *Sutherlandin C* and *Sutherlandin D*. Flavonoids occur in a free state and as glycosides. These groups of constituents are the largest group of naturally occurring phenols (Evans, 2002). Flavonoids are known for their anti-inflammatory and anti-allergic effects, for anti-thrombotic and vaso-protective properties, for inhibition of tumour promotion and as a protective barrier for the gastric mucosa (Evans, 2002).

2.5.2.4 Pinitol

S. frutescens also contains pinitol, which is one of three principle constituents of *S. frutescens*, and a known anti-diabetic agent that may have an application in treating diabetes and AIDS (Ostlund & Sherman, 1996). Pinitol is a type of sugar found in many legumes and is classified as chiro-inositol (Mills *et al.*, 2005), and has been recognized for its anti-diabetic and immune-modulating effects by interacting with dendritic cell maturation (Catelani *et al.*, 2008). Pinitol exerts an insulin-like effect resulting in a decreased level of blood glucose, hence an increased availability of glucose for cell metabolism (van Wyk & Albrecht, 2008). Kim *et al.*, (2007) evaluated the effect of pinitol therapy in T2DM patients who were poorly controlled on hypoglycaemic drugs. After 12 weeks on pinitol treatment (20 mg/kg daily), fasting glucose and post-prandial glucose levels were significantly decreased in T2DM patients indicating that pinitol treatment can alter glucose metabolism.

2.5.2.5 Triterpenoid saponins and other compounds

A minimum of 56 different triterpene glycosides has been found in *S. frutescens* (van Wyk & Albrecht, 2008). *S. frutescens* leaves also contain high levels of unidentified polysaccharides (van Wyk & Albrecht, 2008).

2.5.3 Uses and pharmacological effects

There are many traditional uses for *S. frutescens* including the treatment of poor appetite, indigestion, stomach complaints, dysentery, colds, influenza, kidney conditions, fever, internal cancers, uterine troubles, liver conditions, backache, rheumatoid arthritis, urinary tract infections, stress and anxiety, dropsy, heart failure and diabetes (van Wyk & Albrecht, 2008).

2.5.3.1 *Sutherlandia frutescens* in diabetes management

Moshe (1998) and van Wyk & Gericke *et al.*, (2000) proposed that the high levels of pinitol in *S. frutescens* leaves is a plausible rationale behind the traditional anti-diabetic use. A review of the limited available pharmacological evidence was presented by Sia (2004), who argued that L-canavanine, other amino acids such as L-arginine and pinitol may contribute to anti-diabetic effects, either directly or via anti-inflammatory and NO inhibitory activity. The anti-inflammatory and NO-inhibitory activity of *S. frutescens* extracts could counteract the insulinitis of autoimmune diabetes by protecting pancreatic beta-cells against reactive oxygen radicals of which NO could be one.

Bates, Jones and Bailey (2000) have shown that pinitol exert an insulin-like effect by reducing blood-sugar levels in diabetic mice. However, a preliminary study of the clinical benefits of pinitol in obese and mild T2DM individuals showed disappointing results (Davies *et al.*, 2000). Ojewole (2004) presented evidence that *S. frutescens* extracts can reduce glucose uptake in streptozotocin (STZ)-treated mice. Chadwick *et al.* (2007) showed statistically significant increases in glucose uptake and no weight gain in pre-diabetic rats receiving *S. frutescens* via their drinking water.

In a study on vervet monkeys (*chlorocebus aethiops*) conducted by the South African Medical Research Council (MRC) (Seier, et al., 2002), it was reported that up to nine times the recommended dose of *Lessertia (L.) frutescens* (81 mg/kg body weight per day for three months) presented no significant changes in relevant haematologic, biochemical and physiological parameters. In conclusion, it was found that *L. frutescens* are non-toxic in vervet monkeys (www.sahealthinfo.org/traditionalmeds/firststudy.htm. 2002).

Results of Phase I of a double-blind placebo-controlled study conducted by The International Center for Indigenous Phytotherapy Studies through the South African Herbal Science and Medicine Institute, University of the Western Cape have just been made available (Johnson, Syce, Nell, Rudeen, & Folk, 2007). In this double-blind placebo randomized controlled study,

the safety of *L. frutescens* in 25 healthy adult volunteers was evaluated. Twelve healthy subjects were selected for the treatment arm and received 400 mg *L. frutescens* leaf powder capsules twice a day. In the control arm, 13 subjects received an identical placebo capsule twice a day. Participants were seen at monthly intervals throughout the 3-month study period. Blood and urine samples for haematology/biochemistry (i.e., Read Blood Cell (RBC), White Blood Cell (WBC), serum analysis and serum proteins) and urinalysis (i.e., Serum Glutamate (SG), Potential of hydrogen (pH), protein, glucose, ketone, bilirubin, blood, & urobilinogen) were measured. Participants also completed a 12-lead Electrocardiogram (ECG) and reported any adverse events at each visit. Results indicate that the 800 mg daily dosage was well tolerated by study participants, with no significant changes present in the relevant haematologic, biochemical or physiological parameters (Johnson *et al.*, 2007).

The National Center for Complementary and Alternative Medicine, National Institutes of Health (NIH) and Department of Medicine at Edendale Hospital, Pietermaritzburg, South Africa evaluated the efficacy of *L. frutescens* in HIV patients (Wilson *et al.*, 2015). In Stage 1, 56 participants were randomized to *S. frutescens* 400, 800 or 1,200 mg twice daily or matching placebo for 24 weeks. In Stage 2, 77 additional participants were randomized to either 1,200 mg *S. frutescens* or placebo. In the final analysis, data from Stage 1 and Stage, 2 were combined such that 107 participants were analysed (54 in the *S. frutescens* 1,200 mg arm and 53 in the placebo arm). *S. frutescens* did not change the HIV viral load, and CD4 T-lymphocyte count was similar in the two arms at 24 weeks. However, mean and total burden of infection (BOI; defined as days of infection-related events in each participant) was greater in the *S. frutescens* arm: mean (SD) 5.0 (5.5) vs. 9.0 (12.7) days ($p=0.045$), attributed to two tuberculosis cases in subjects taking isoniazid preventive therapy (IPT), Wilson *et al.*, (2015).

2.5.4 Toxicology

S. frutescens has a long history of traditional use, with no reports of any serious side effects, suggesting that it can be considered as generally safe. As stated by Mills *et al.* (2005) with recorded side effects as dryness of mouth, occasional mild diarrhea or mild diuresis and dizziness in cachectic patients. Infusions or decoctions of 2.5 to 5 g of dry material per day can be regarded as the traditional dose (van Wyk *et al.*, 1997). For commercial preparations, 300 mg of dried leaves twice daily (i.e. 600 mg per day) is recommended (with the usual precaution that it should be avoided during pregnancy or lactation). According to Johnson *et*

al., (2007) in Phase I of a safety clinical study on 25 healthy adults at Karl Bremer Hospital, Bellville, South Africa, showed that two 400 mg leaf powder capsules (800 mg per day) was well tolerated, with no side effects noted during or after the three months' trial period. There was no change in frequency of adverse events, or any clinically significant changes in most physical, vital, blood and biomarker indices. The possibility exists that products containing *S. frutescence* may interact with antiretroviral medication (Mills *et al.*, 2005) or with insulin and other diabetes medication (Sia, 2004).

2.6 Clinical trials

While new clinical trials on herbal medicine are being published at an increasing rate, there are still huge gaps in the evidence base; however, most of these studies are not considered sufficiently rigorous (Firenzuoli & Gori, 2007). A survey of the specialized literature reveals that few well-controlled trials have been carried out with herbal medicines (Bansal, 2010). A recent meta-analysis of reviews published in important medical journals, such as the *Annals of Internal Medicine*, the *Journal of the American Medical Association (JAMA)*, the *British Medical Journal*, the *Lancet*, and the *British Journal of Clinical Pharmacology*, among others, confirms this assumption (Bansal, 2010). The lack of pharmacological and clinical data on the majority of herbal medicinal products is a major impediment to the integration of herbal medicines into conventional medical practices.

The main question that often has not been answered satisfactorily, deals with the triad absorption, metabolism and efficacy of herbs and their extracts, and is actually an important unsolved problem in judging their many alleged health effects (Firenzuoli, 2006).

Evaluation of the efficacy of herbal products and applying the principles of modern medicine is a paramount issue. The “gold standard” of evidence for treatment efficacy within evidence-based medicine (EBM) is considered to be a systematic review or meta-analysis of randomized controlled trials (RCT) with double blinding and a comparator group (Firenzuoli & Gori, 2007). The evaluation of herbal products with similar principles of EBM poses a certain unique issue (Gagnier *et al.*, 2006). The (CONSORT) statement for trials of herbal medicines can be a very important paradigm to follow in evaluating the efficacy, effectiveness and safety of traditional herbal remedies. Five criteria is recommended: quantity and quality of available preliminary data to help determine the most appropriate type of research; extent of use by the public; public

health importance of the disease being treated; the feasibility of conducting the research; and the cost of the research.

2.7 Regulatory control of herbal medicine

The future of herbal drugs is overshadowed by the pervading lack of regulatory control. The diversity among countries with a long history and holistic approach of herbal medicines makes evaluating and regulating them very challenging. In addition, there are a great number of different herbs used. Legislative criteria to establish traditionally used herbal medicines as part of an approved health care faces several difficulties. In a survey conducted across 129 countries, the WHO reported the following issues regarding herbal medicines: lack of research data, appropriate mechanisms for control of herbal medicines, education and training, expertise within the national health authorities and control agency, information sharing, safety monitoring and methods to evaluate their safety and efficacy. The support needed from different countries includes information sharing on regulatory issues, workshops on safety monitoring of herbal medicines, general guidelines on research and evaluation of herbal medicines, provision of databases, herbal medicine regulation workshops and international meetings (WHO, 2005).

National policies are the basis for defining the role of TMs in national healthcare programmes, ensuring that the necessary regulatory and legal mechanisms are established for promoting and maintaining good practice, assuring the authenticity, safety, and efficacy of TMs and therapies and providing equitable access to health-care resources and their resource information (WHO, 2005). Another fundamental requirement is harmonization of the market for herbal medicines for industry, health professionals and consumers (Mahady, 2001). The definition and categorization of herbal medicines vary from one country to another. Depending on the regulations which apply to foods and medicines, a single medicinal plant may be categorized as a food, a functional food, a dietary supplement, or herbal medicine in different countries. A common regulatory framework does not exist in different countries. As a result, information on clinical indications for their use, efficacy and safety are influenced by the traditional experience available in each place (Wachtel-Galor & Benzie, 2011).

In the United States of America (USA), for example, the Dietary Supplement Health and Education Act (DSHEA) of 1994 classify herbs as dietary supplements. Supplements are defined broadly as “anything that supplements the diet”. A major difference between a drug

and a dietary supplement is that dietary supplements may not claim to “diagnose, cure, mitigate, treat, or prevent illness”. Dietary supplements can be produced, sold, and marketed without first demonstrating safety and efficacy, as is required for pharmaceutical drugs. Also, the Food and Drug Administration (FDA) bears the regulatory burden of proving that a dietary supplement is unsafe before it can be removed from the market which is in direct contrast to drugs, where a manufacturer must provide the FDA with evidence of safety and efficacy before a product can be sold (Bent, 2008).

In Europe, the European Directive 2004/24/EC, released in 2004 by the European Parliament and by the Council of Europe, provides guidelines for the use of herbal medicines (Calapai, 2008). The directive establishes that herbal medicines released on the market need authorization by the national regulatory authorities of each European country and that these products must have a recognized level of safety and efficacy (Calapai, 2008). The registration of herbal medicinal products needs sufficient evidence for the medicinal use of the product throughout a period of at least 30 years in the European Union (EU), at least 15 years within the EU, and 15 years elsewhere for products from outside the EU (Routledge, 2008; Vlietinck, Pieters, & Apers, 2009).

These major differences among countries introduce serious difficulty in the definition of the concept of herbal medicines for the purposes of national drug regulation, while simultaneously, is also confusing patients and consumers (WHO, 2005). The WHO has published guidelines to define basic criteria for evaluating the quality, safety and efficacy of herbal medicines aimed at assisting national regulatory authorities, scientific organizations and manufacturers in this particular area (Wachtel-Galor & Benzie, 2011). Furthermore, the WHO has prepared pharmacopoeia monographs on herbal medicines and the basis of guidelines for the assessment of herbal drugs (Calixto, 2000; Zhang, 1998).

2.8 Ethics of herbal medicine

An ethical framework for clinical research has two objectives, which is to promote socially valuable clinical investigation and to protect research subjects from exploitation. Emanuel, Wendler & Grady 2000 and Emanuel & Miller *et al.*, (2001) proposed a framework consisting of requirements that must be satisfied for ethical clinical research. First, the research should have social value, that is, it should generate knowledge that leads to improved health and health care. Second, the clinical research should have scientific validity and not expose subjects to

risk without the potential to produce generalizable knowledge. Third, clinical research must select subjects fairly in accordance with the scientific objectives of the study and avoid unnecessary involvement of vulnerable groups. Fourth, all clinical research must have a favourable risk-benefit ratio, minimizing risks to subjects and justifying the risks by the potential benefits to subjects and the value of the knowledge to be gained from the research. Fifth, to protect subjects and ensure public accountability, all clinical research studies should receive a prospective and ongoing review by a committee composed of individuals independent of the research. Sixth, competent adults should not be enrolled in research unless they have been adequately informed about the study and they have agreed to participate. For research with children and incompetent adults, informed authorization by parents or other surrogate decision makers is required. Seventh, research must be conducted in a way that respects the rights and protects the well-being of enrolled subjects. Because herbal medicine historically has lacked the established research infrastructure of conventional medicine, it has attracted relatively few high calibre researchers (Emanuel *et al.*, 2004 & Tilburt *et al.*, 2008). As a consequence, the field suffers from a general lack of research expertise (potentiated and perpetuated by lack of funds). This, in turn, has resulted in a situation where many of the relatively few scientific investigations in herbal medicine are methodologically weak or outright flawed. Yet flawed science is unlikely to be ethical, expressed in the words of the British Medical Association, “Studies which are unscientific are also unethical” (Ernst, 2003). According to the Guidelines for Good Practice in the Conduct of Clinical Trials with Human Participants in South Africa (Department of Health, 2006), “all clinical trials conducted in South Africa must undergo ethical review by an accredited ethics committee”. The main responsibility of Research Ethics Committee REC in South Africa is to ensure the protection of, and respect the rights, safety and well-being of participants involved in a trial and to provide public assurance of that protection by reviewing, approving and providing comment on clinical trial protocols, the suitability of investigator(s), facilities, methods and procedures used to obtain informed consent as ethical review affects the potential participants and the general day to day functioning of the health system (Department of Health, 2006). The committee is required to be accredited by the National Health Research Ethics Council.

In summary, type 2 diabetes is the most common form of diabetes, accounting for 90–95% of cases (Centers for Disease Control, 2011). Furthermore, it has been estimated that diabetes

prevalence for South Africans is at 6.46% for adults, age 20–79 years. T2DM is associated with serious long-term microvascular and macrovascular complications such as cardiovascular disease, neuropathy and diseases of the eyes, which in turn, increase morbidity, mortality and healthcare costs. Although there is no cure for T2DM, *S. frutescens* is an old Cape remedy which is used for various ailments. Traditionally, this includes T2DM, since *S. frutescens* has been shown to have anti-diabetic properties and has shown to be safe in healthy volunteers. However, no clinical study has yet been done to test the efficacy of *S. frutescens* in T2DM. Therefore, a protocol will be designed according to guidelines and Good Clinical Practice (GCP) of South Africa, for testing the efficacy of *S. frutescens* in adult T2DM including the preparation of an application to the MCC and ethics committee.



UNIVERSITY of the
WESTERN CAPE

Chapter 3

Plan of Work

In this chapter, the objectives set for this study, the hypotheses to be tested and the study approach is presented.

3.1 Study objectives

The objectives of the study were to:

1. Establish a scientifically sound rationale for a clinical study to evaluate the efficacy of *Sutherlandia frutescens* (*S. frutescens*) in adult type 2 diabetes mellitus patients (T2DM).
2. Design and develop a clinical trial protocol for a Phase II efficacy study of *S. frutescens* in adult T2DM.
3. Prepare an application document for the regulatory approval of an efficacy trial of *S. frutescens* in adult T2DM and
4. Prepare an application document for ethics approval of an efficacy trial of *S. frutescens* in adult T2DM.

3.2 Hypotheses

It was hypothesized that:

- *S. frutescens* may have anti-diabetic properties and therefore may be evaluated for its anti-diabetic efficacy in adults with T2DM.
- A protocol and regulatory authority and ethics application documents required for successfully obtaining approval from the Medicine Control Council (MCC) and a relevant South African ethics committee (EC) for a trial of *S. frutescens* in adult diabetics should be relatively easy to be developed.

3.3 Study approach

To realize the above objectives the following were done.

3.3.1 Determination of a scientific rationale to test *Sutherlandia frutescens* in type 2 diabetes mellitus

To realize this objective, a search was done for available medical literature on *S. frutescens* and clinical trials on herbal medicines, specifically those used for the treatment of T2DM. The clinical trials done on herbal medicine for T2DM were reviewed to gain insight into the overall efficacy, if any, of the products and to determine the number and type of previous clinical studies on herbal medicine in T2DM. These were further reviewed and assessed for the therapeutic outcomes, type of patient, study design, potential active ingredients and the challenges typically involved.

Searches were done to retrieve all information on *S. frutescens* and trials done on herbal medicine as anti-diabetic drugs. These searches were achieved in the first semester of 2015 and the search terms used included: clinical trials alternative medicine, clinical trials traditional herbal medicine, clinical trials herbal medicine anti-diabetics, *S. frutescens*, T2DM and randomized controlled trial. The websites used for these searches included: www.clinicaltrials.gov; www.google.com; www.PubMed.com; Science direct and Google Scholar. The information collected was read and analyzed for the number and type of studies which could be located for a sound rationale for a clinical study of *S. frutescens*.

3.3.2 Design and development of a clinical trial protocol

To achieve the second objective, a protocol was developed to evaluate the efficacy of the herbal preparation, *S. frutescens*, to treat adults with T2DM by means of a clinical study. This plant is known to be widely used for diabetes (Sia, C. 2004) and the clinical evaluation thereof would certainly prove to be valuable. Using the information prepared in the (Chapter 4) i.e. trials on herbal medicines and reported trials already conducted on herbal medicines, a protocol for an efficacy study on *S. frutescens* was written. In this protocol, specific attention was given to the following : the study design; the study objective; endpoints; subjects; inclusion and exclusion criteria; the trial material; statistical analysis; patient information and consent forms. The protocol had to be executable, meet the criteria for acceptance by the ethics committees and regulatory authority, be useful to recruit volunteers and be acceptable to trial investigators. The

protocol to be designed and developed had to be able to test the anti-diabetic effect of the herbal plant *S. frutescens* in diabetic subjects.

For the purpose of this investigation, it was decided to design a 24-week, randomized, double blind study, with the endpoints being, efficacy and safety. Exclusion and inclusion criteria were considered on the grounds of gender, age, disease risk factors, medical history, stage of disease and concomitant medication. To ensure that the study was of a high standard, it was preferable for the study to be randomized, placebo controlled and double-blinded. The quality and dosage form of the herbal preparation, *S. frutescens*, was also a very important aspect of the study design. Other reference trials and a statistician were consulted to argue about what meaningful change would be regarded as the primary endpoint produced by the herbal plant in comparison to that of the placebo.

Finally, a patient information leaflet and consent form were designed to accompany the protocol and to adequately inform the volunteers of the aims, methods, sources of funding, potential hazards or discomforts the study may entail and their freedom to withdraw consent.

3.3.3 Preparation of the application document for regulatory approval

For the third objective, to prepare the application document required for regulatory approval of an efficacy trial of *S. frutescens* in treating adult T2DM, the MCC specifications had to be followed. Application forms for submission were obtained from this regulatory authority via the internet at www.mccza.com. All the necessary completed forms as well as copies of the protocol, patient information leaflet, a covering letter, insurance details and recruitment advertisements were prepared, ready for submission to the Registrar in Pretoria strictly following the MCC submission rules.

3.3.4 Preparation of the application document for ethics approval

An internet search was done to execute the fourth objective, i.e. to prepare the document needed for ethical approval of an efficacy trial of *S. frutescens* in treating adult T2DM. For this, information on the ethics requirements for herbal products was searched and assessed for the criteria that may be used for registering *S. frutescens*. The following key phrases were used: “ethics of herbal products”, “herbal medicines”, “guidelines of herbal medicines” and “requirements for registration of herbal medicines”. Websites accessed were, Google scholar, PubMed, Google, and for ethics application forms, the University of Stellenbosch website at

www.sun.ac.za was accessed. Application forms and guidelines were downloaded from the Health Research Ethics Committee (HREC) tab. The MCC requires that all clinical trials using medicinal products should obtain ethical clearance from an ethics committee that is structured and operates according to Good clinical Practice (GCP) guidelines. Upon completion of the entry form, the application would be ready for submission to the ethics committee for approval. From the information gathered, a list of ethics requirements appropriate for approval and registration of *S. frutescens* capsule herbal product would be available.



UNIVERSITY of the
WESTERN CAPE

Chapter 4

A scientifically sound rationale for testing *Sutherlandia frutescens* in type 2 diabetes mellitus

4.1 Introduction

The South African indigenous medicinal plant, *Sutherlandia frutescens* (*S. frutescens*), has traditionally been used for the treatment of a variety of diseases, including type 2 diabetes (T2DM), i.e. having a potential hypoglycaemic effect. However, no clinical study has yet been done to test the efficacy of *S. frutescens* in T2DM. Hence, a scientifically sound rationale is required for the use of *S. frutescens* in T2DM. The medicinal plant should be tested for efficacy, in a dosage form, on specific patients, in a particular study design (protocol). Consequently, the objective of this part of the thesis was to establish a scientifically sound rationale for testing the efficacy and safety of *S. frutescens* in T2DM.

In this chapter, (i) review previous clinical studies of herbal medicine in the treatment of T2DM will be evaluated (ii) The traditional uses and possible the pharmacologically active compounds of *S. frutescens* will be identified. (iii) Pre-clinical studies of *S. frutescens* in diabetic conditions may suggest reasons for testing this plant as an anti-diabetic and finally, (iv) the safety data and dosage forms of *S. frutescens* will be determined.

The results obtained will be reported, discussed and a final conclusion drawn. Thus, a sound scientific rationale for conducting a clinical study of *S. frutescens* in T2DM is presented.

4.2 Methods

To realise the above stated objective, the following were achieved:

4.2.1 Review previous clinical studies of herbal medicine in the treatment of type 2 diabetes mellitus

The following Internet-based sources viz. Medline, PubMed, Google, Science Direct, Google Scholar, Cochrane Airways Group Specialised Register, Cochrane Complementary Medicine Field Specialised Register, the Cochrane Central Register of Controlled Trials (CENTRAL) and Embase databases were accessed for the relevant information. Among the search terms/keywords used were: clinical trials alternative medicine, clinical trials traditional herbal medicine, clinical trials diabetes and clinical trials herbal medicine diabetes. All relevant information was collected, read and analysed for the number and type of previous clinical studies on herbal medicine in T2DM. These were further reviewed and assessed according to the following variable: (a) objectives of the study (b) type of therapeutic outcomes (c) dosage forms (d) therapeutic parameters measured, (e) study design and (f) study challenges.

4.2.2 The traditional uses and possible pharmacologically active compounds of *Sutherlandia frutescens*

To obtain the information on traditional uses and pharmacological active compounds of *S. frutescens*, a number of search engines were used to access related electronic databases. They include: PubMed, Google, Medline, Science Direct, Cochrane Library Database, Google scholar. The following search terms (or keywords) were used: *Sutherlandia frutescens*, diabetes and traditional herbal medicine. In this section, we listed and summarized the existing traditional uses of *S. frutescens* especially for the treatment of diabetes traditionally and the pharmacological active compounds were also identified which were relevant for T2DM.

4.2.3 Pre-clinical studies of *Sutherlandia frutescens* in diabetes mellitus

Literature searches were performed to identify all published articles on pre-clinical studies of *S. frutescens* in diabetic conditions, especially those that may suggest reasons for testing this plant in T2DM and those suggesting efficacy studies. The main search engines used were (PubMed, and Google, Science Direct, Google Scholar and Medline). All the required articles

with a focus on the study objective, problems and duration, parameter measured and the active compounds were reviewed and analysed and the results were presented and discussed.

4.2.4 The safety data and dosage forms of *Sutherlandia frutescens*

A similar Internet search as in 4.2.3 was done to retrieve the information on *S. frutescens*, to identify the articles that focused on the safety data and dosage formulation (e.g. tablet or capsule) versus the active content of *S. frutescens*.

4.3 Results and discussion

4.3.1 Review of previous clinical studies of herbal medicine in the treatment of type 2 diabetes mellitus

The extensive search on the literature for clinical trials on herbal medicines lead to more than 20 review articles, out of which we identified specific articles from seven studies. These clinical trials were performed in different countries and varied substantially. We included these studies as randomized clinical trials which had been done on herbal treatments for anti-diabetic patients and presented as summary in table 4.1. We discuss table 4.1 hereafter. (a) Objectives of the study (b) type of therapeutic outcomes (c) dosage forms (d) therapeutic parameters measured, (e) study design and (f) study challenges

Table 4. 1: Clinical studies of herbal medicines for type 2 diabetes mellitus

Plant (scientific name) Authors (years)	Objective	Outcome	Dosage forms	Parameters measured	Study design	Duration & Number of patients	Study challenges
<i>Ayurvedic formulation</i> Awasthi et al. (2015)	To investigate the efficacy of a standardized polyherbal formulation consists of aqueous extracts from six herbs, in patients with T2DM	↓fasting and post prandial blood glucose, ↓glycated haemoglobin (HbA1c), ↓total cholesterol level	polyherbal capsules 500 mg/day +Metformin 500 mg/day	Blood glucose and HbA1c & Lipid levels	Randomized control study (RCT)	6 months / 93 patients	The six months duration of the study was sufficient to observe the changes in the biochemical parameters but also short enough to emphasize the side effects of study drugs in the long term. Although this study lacked a placebo control group due to ethical reason.
<i>Allium cepa (A. cepa),</i> Myint et al. (2009)	To investigate the hypoglycemic effects of <i>Allium cepa</i> in patients with T2DM	↓fasting plasma glucose level significantly reduced the induced hyperglycaemia (GTT)	100 mg , <i>Allium cepa</i>	FBG, glucose level	Controlled study	16 weeks / 21 patients	Was evident that, <i>Allium cepa</i> has hypoglycemic effects that may be beneficial in management of diabetes in addition to its other nutritional values.
<i>Salacia reticulata</i> Jayawardena et al. (2005)	Investigate the effects of a herbal tea containing <i>Salacia reticulata</i> (Kothala Himbutu tea) in patients with T2DM	↓HbA1c, ↓ fasting blood glucose (FBG)	Standard tea bags	Blood glucose and HbA1c7& FBG	Randomized single-center double-blind crossover	6 weeks / 28/23 patients	It was significant falls in HbA1C levels. In addition, it lowered the required dose of glibenclamide. The drug was well tolerated with comparable proportions of patients in placebo and drug treatment groups experiencing adverse effects
<i>Trigonella foenum-graecum</i> Lu et al. (2008)	To evaluate the efficacy and safety of (TFGs) in combination with sulfonylureas (SU) in the treatment of patients	↓fasting blood glucose, ↓HbA1c	10.2 g/day	PBG, HbA1c	RCT double-blind	12 weeks / 46 patients	The combined therapy of TFGs with sulfonylureas hypoglycemic drug could lower the blood glucose level and ameliorate clinical symptoms in the treatment of T2DM, and the therapy was relatively safe.
<i>Silybum Marianum</i> Huseini et al. (2006)	T2DM	↓fasting blood glucose, ↓LDL, ↓ cholesterol, ↓TG, ↓SGOT, ↓ SGPT	600 mg+ Standard therapy	FBG, Cholesterol level, SGOT, SGPT	RCT	4 months / 29/25 patients	silymarin treatment in type II diabetic patients for 4 months has a beneficial effect on improving the glycemic profile
<i>Cinnamon</i> Mang et al. (2006)	Aqueous <i>cinnamon</i> purified extract improves glycated haemoglobin A1c(HA1c)	No adverse effects were observed, ↓fasting plasma glucose	3 g of cinnamon powder per day	Fasting plasma glucose & HbA1c& Total cholesterol	Randomized double-blind placebo control	4 months / 79 patients	cinnamon extract seems to have a moderate effect in reducing fasting plasma glucose concentrations in diabetic patients with poor glycaemic control
<i>Juglans regia</i> Hosseini et al. (2014)	To investigate the effects of the <i>Juglans regia</i> leaf extract on hyperglycemia and lipid profiles in T2DM	↓HbA1c, ↓FBG, ↓total cholesterol and triglyceride level. No live and kidney and other side effect were observed	100 mg	FBG, HbA1c, triglyceride & totalcholesterol	Randomized double-blind placebo control	3 months / 61 patients	The three months duration of the study was significantly decrease FBG, HbA1c, triglyceride and totalcholesterol of type II diabetic patients without important adverse effects.

1. *Ayurvedic formulation*

In a six-month randomized, active-controlled study, researchers investigated newly diagnosed T2DM participants. The expected outcomes primarily measured the effective change that an Ayurvedic formulation had on baseline blood glucose (fasting blood glucose and postprandial blood glucose) and glycosylated haemoglobin (HbA1c). A secondary outcome involved the effect that this treatment had on lipid levels, liver enzymes and renal function test (Awasthi *et al.*, 2015). A random sample of 93 newly diagnosed T2DM patients were divided into group 1, who received polyherbal formulation (PHF) capsules 500 mg/day, up-titrated weekly to a maximum of 3 g/day; while group 2 received metformin 500 mg/day, up-titrated weekly to a maximum of 2 g/day.

Mean fasting and postprandial blood glucose (PPBG) of group 1 (on PHF) showed a decrease of 25.52% and 24.22%, respectively, in comparison to 31.46% and 24% in group 2 (on metformin) after 24 weeks. The estimated treatment difference was -10.8; 95% CI -22.63 to 1.03 and -0.36; -12.1 to 11.38, respectively. A similar reduction in HbA1c was noted in both groups, with an estimated treatment difference of 0.01; 95% CI -0.51 to 0.53. Mean total cholesterol level, on the other hand, was markedly decreased in group 1 (estimated mean difference 61.3; 95% CI 55.32 to 67.28) than in group 2 (estimated mean difference 41.12; 95% CI 34.92 to 47.32). At the end of six months there was a statistically significant difference between the treatment groups in their total cholesterol level (estimated treatment difference 20.18; 95% CI 12.34 to 28.02. (Awasthi *et al.*, 2015).

2. *Allium cepa (A. cepa)*

Mathew and Augusti (1974) reported that oral consumption of *Allium cepa* (onion) can improve glycaemic control in diabetes. More recently, an intake of 100 g *A. cepa* was shown to decrease the fasting blood glucose (FBG) level and improve glucose tolerance (GTT) in T1DM and T2DM patients (Eldin *et al.*, 2009). In a self-controlled study on 21 T2DM patients, an acute hypoglycaemic effect of *A. cepa* was also observed, while an attenuated (37%) rise in plasma glucose was found two hours after glucose ingestion (Myint *et al.*, 2009).

3. *Salacia reticulata (S. reticulata)*

A diet containing aqueous extract from the stem of *Salacia reticulata* (240 mg/day for 6 weeks) was shown to decrease FBG and HbA1c levels in T2DM patients (Kajimoto *et al.*, 2000). A significant reduction in HbA1c was also reported in the patients receiving a preparation of *S.*

reticulate tea for three months (Jayawardena *et al.*, 2005). Clinical efficacy of *S. reticulate* consumption (2 g/day for 3 months) in the management of diabetes had also been observed in 30 patients (Radha & Amrithaveni, 2009).

4. *Trigonella foenum-graecum* (*T. foenum-graecum*)

The hypoglycaemic effect of *Trigonella foenum-graecum* (fenugreek) seeds has been demonstrated in cell culture, animal models and humans with more than 30 studies (Ghorbani & Rakhshandeh, 2012). In a double-blind placebo controlled trial, 46 T2DM patients were given sulfonylureas drug plus *T. foenum-graecum* seeds (in tablet form; 6 tablets/3 times a day) or sulfonylurea drug plus placebo (23 cases). After 12 weeks, the combined therapy had more effect on the level of FBG, HbA1c, and PPBG. (Lu *et al.*, 2008).

5. *Silybum marianum* (*S. marianum*)

In a 4-month randomized double-blind clinical study, *Silymarin* (200 mg 3 times a day) could decrease FBG, HbA1c, total cholesterol, LDL, TG, serum glutamic oxalacetic transaminase (SGOT) and serum glutamic pyruvic transaminase (SGPT) in T2DM patients (30 cases) receiving conventional therapy (Huseini *et al.*, 2006). A reduction in glucose, lipids and hepatic enzymes is consistent with the results of another trial (Hussain *et al.*, 2007; Ramezani *et al.*, 2008), among 25 T2DM patients receiving the same dose of *silymarin* for four months (Huseini *et al.*, 2006). Beneficial effects of *silymarin* (200 mg 3/day) on FBG, HbA1c and PPBG have also been seen in T2DM patients maintained on glibenclamide (Huseini *et al.*, 2006).

6. *Cinnamon*

In a double-blind study over four months, 79 patients with diagnosed T2DM who were treated with oral anti-diabetics were randomly assigned to take either a *cinnamon extract* or a placebo capsule three times a day (Mang *et al.*, 2006). The amount of aqueous *cinnamon extract* corresponded to 3 g *cinnamon powder* per day. There was a significantly higher reduction in the *cinnamon* group (10.3%) than in the placebo group (3.4%). No significant intragroup or intergroup differences were observed regarding HbA1c, lipid profiles or differences between the pre- and post-intervention levels of these variables. The decrease in plasma glucose correlated significantly with the baseline concentrations, indicating that subjects with a higher initial plasma glucose level may benefit more from the *cinnamon* intake. No adverse effects were observed (Mang *et al.*, 2006).

7. *Juglans regia*

A 3-month randomized double-blind, placebo-controlled clinical trial was conducted on 61 T2DM patients, aged 40–60 years, with FBG between 150 mg/dL and 200 mg/dL, HbA1c between 7% and 9%. They were divided into two groups. The first group received 100 mg *Juglans regia* leaf extract in a capsule twice daily for three months, while the other group received a 100 mg placebo capsule with the same dosage. The standard anti-diabetic therapy (metformin and glibenclamide, and nutritional regimen) was continued in both groups. At baseline and after three months, the FBG, insulin, HbA1c, cholesterol, triglyceride, HDL, LDL levels, liver and renal function tests were determined. In addition, general satisfaction with the treatment was identified using health questionnaires (Hosseini *et al.*, 2014). Results showed that FBG, HbA1c, total cholesterol and triglyceride levels in patients treated with *Juglans regia* significantly decreased compared with their baseline and the placebo group. These patients were also significantly satisfied with *Juglans regia* treatment compared with the placebo group. No liver or kidney abnormalities or other side effects were encountered in the groups. However, some GI events (especially mild diarrhoea) were linked to the extract treatment at the start of the study. (Hosseini *et al.*, 2014).

4.3.2 The traditional uses of *Sutherlandia frutescens* and its pharmacological active compounds

In South Africa, *S. frutescens* is used by an array of healers, such as herbalists. It has been used for variety of disease that includes diabetes. The aerial parts of the plant (stems, leaves, flowers, and pods), the roots, or only the leaves are usually used to make the infusions and decoctions. A decoction of *S. frutescens* is used to wash wounds and the eyes and to reduce fevers (Van Wyk & van Staden 2002) and the infusions from the leaves and stems are used to treat diabetes, and other ailments.

Traditional healers prepare the decoctions according to the type of disease indicated by the patient. Their preparation ranges from using the leaves, flowers, stems, and roots for different ailments (Aboyade & Hughes 2014). The traditional healers collect fresh plant materials, dry them and then stamp or mash them into powder between two stones. The preparation is then usually infused in hot or boiling water, left to cool and given to the sick person.

Traditionally, the plant is used in liquid dosage form such as tea and decoctions. Currently, *S. frutescens* is available in various dosage forms, such as capsules and tablets (containing *S.*

frutescens raw material in powdered form), gels for topical application, creams, liquid extracts, and ointments (Van Wyk & Albrecht 2008) and is found in pharmacies and herbal shops.

The mechanism of action of *S. frutescens* for these disease and conditions is not known, but several attempts are being made to understand how this plant works. Various chemical compounds that might be responsible for the activity of *S. frutescens* have been identified. These include triterpenoids, saponins, flavonoids, γ -aminobutyric acid (GABA) and pinitol (Van Wyk & Albrecht, 2008). The latter compound may be responsible for the antidiabetic effects and has been found to possess insulin-like properties (Bates, Jones. & Bailey, 2000). It promotes stimulation of glucose uptake as well as translocation of the glucose transporter 4 (GLUT4) to the plasma membrane (Dang & Ashida, 2010). Hence, the other active ingredients might be useful for such conditions.

4.3.3 Pre-clinical studies of *Sutherlandia frutescens* in diabetes mellitus

Internet searches were done and limited studies were found on *S. frutescens* in diabetes. Seven studies were identified, presented and discussed.

Moshe (1998) and van Wyk *et al.*, (2000) proposed that the high levels of pinitol in *S. frutescens* leaves is a plausible rationale behind its traditional anti-diabetic use. A review of the limited available pharmacological evidence was presented by Sia (2004), who argued that L-canavanine, other amino acids such as L-arginine and pinitol may contribute to the anti-diabetic effects, either directly or via anti-inflammatory and nitric oxide inhibitory activity. The anti-inflammatory and nitric oxide -inhibitory activity of *S. frutescens* extracts could counteract the insulinitis of autoimmune diabetes by protecting pancreatic beta-cells against reactive oxygen radicals of which nitric oxide could be one. Bates, Jones and Bailey (2000) have shown that pinitol exert an insulin-like effect by reducing blood sugar levels in diabetic mice, however, a preliminary study of the clinical benefits of pinitol in obese and mild type 2 diabetic individuals showed disappointing results (Davies *et al.*, 2000). Ojewole (2004) presented evidence that *S. frutescens* extracts can reduce glucose uptake in STZ-treated mice.

Chadwick *et al.* (2007) investigated the hypoglycaemic effects of *S. frutescens* in diabetic rats. Wistar rats received a diet specifically designed to induce obesity, insulin resistance and leading to a typical prediabetes state. The rats were then divided into three groups. The first group received metformin. Metformin is a drug administered to hyperglycaemic individuals. The second group of rats received an infusion made with the leaves of *S. frutescens*, and the

third group of rats served as a control. After 8 weeks of medicinal compliance, the rats receiving *S. frutescens* in their drinking water displayed normal insulin levels. There was a marked increase in glucose uptake into muscles and adipose tissue and a significant decrease in intestinal glucose uptake. The results obtained from this experiment indicate that *S. frutescens* extract has beneficial effects on T2DM.

Williams *et al.* (2013) investigated the capacity of an aqueous extract of *S. frutescens* to prevent insulin resistance (a precursor to type 2 diabetes) in a human liver cell culture and to identify genes regulated by *S. frutescens* treatment. In their study they found that *S. frutescens* could prevent insulin resistance in hepatocytes. Changes in gene expression indicated several potential mechanisms of anti-diabetic action for *S. frutescens*, reflecting the multiple bioactive compounds previously identified in aqueous extracts of *S. frutescens*.

S. frutescens contains pinitol, which is a known anti-diabetic agent that may have an application in treating wasting in diabetics (Ostlund & Sherman, 1996). *S. frutescens* extracts show promise as a medication for type 2 diabetes, but the clinical efficacy and mechanism(s) of action need further study (Chadwick *et al.*, 2007).

4.3.4 Safety data and dosage forms of *Sutherlandia frutescens*

Internet searches that were done yielded two articles on the safety of *S. frutescens*. No serious adverse effects have been reported since ancient times. However, a scientific study recommended a daily dose of the *S. frutescens* leaf powder in a detailed first study on the safety of the therapeutic dose in vervet monkeys. Seier *et al.* (2002) reported that they were given 0, 1, 3 and 9 times the recommended daily dose of 9.0 mg/kg body weight (i.e. 0, 9.0, 27.0 and 81.0 mg of leaf powder). This was administered as part of a carefully monitored standard diet for a period of three months. No clinically significant toxic- or side effects were observed in a detailed evaluation of 15 haematological, 21 clinical, biochemical, six physiological and several behavioural variables (Seier *et al.*, 2002). The dried, ground herb was infused in one litre of boiling water and cooled. The infusion was then strained and taken in half tea-cup doses (90 ml) three times daily (Matsabisa, 2006; Mills *et al.*, 2005). Children 6–12 years: one-quarter tea cup (45 ml) three times daily (Matsabisa, 2006). A recent Phase I clinical study (healthy volunteers) by Johnson *et al.*, (2007), (as mentioned in Chapter 2 points 2.5.3.1) showed a statistically significant increase in appetite in the treatment group as well as a lower respiration rate ($P < 0.04$), a higher platelet count ($P < 0.03$), MCH ($P < 0.01$), MCHC ($P < 0.02$),

total protein ($P < 0.03$) and albumin levels ($P < 0.03$). These differences remained within the normal physiological range and were not considered clinically relevant

4.4 Conclusions

based on the information gathered from the literature reported, experience on clinical herbal medicine studies and claims on their traditional uses, safety and efficacy is not adequately supported by clinical evidence. *S. frutescens* has been used traditionally in diabetes and has anti-diabetic properties. The primary active compounds found in *S. frutescens* may possibly be acting synergistically in the human organism, mediating greater clinical benefit than noticed in single compound studies. Consequently, studies on *S. frutescens* has shown to be non-toxic, with no adverse or side effects and is reportedly safe in healthy volunteers. Experiences on similar herbal products clinically tested in diabetics or other disease states.

Overall, based on information on the traditional use, chemistry and results of existing pre-clinical and clinical safety studies of *S. frutescens*, as well as literature reported experiences on similar herbal products clinically tested in diabetes, a sound rationale for testing the efficacy of *S. frutescens* in adult type 2 diabetes patients was successfully established.



UNIVERSITY of the
WESTERN CAPE

Chapter 5

Design and development of a clinical trial protocol to determine the efficacy of

Sutherlandia frutescens in adult type 2 diabetes mellitus

5.1 Introduction

Type 2 diabetes mellitus (T2DM) is one of the endocrine disorders and metabolic with a high prevalence and one which can be treated. Characteristics which distinguish T2DM are hyperglycaemia, insulin resistance and a relative lack of insulin secretion, which occurs in as much as 90% of all cases (Soltani, Gorji, Asgary, Sarrafzadegan & Siavash, 2015). According to the World Health Organization (WHO) and International Diabetes Federation (IDF), the number of people affected with diabetes has increased dramatically over recent years, as has the prevalence globally (WHO, 2011). Currently, there are over 366 million diabetics worldwide and this is likely to increase to 552 million by the year 2030 (Whiting *et al.*, 2011; WHO, 2011). Though there is a high prevalence, treatment is available for T2DM. The condition can be controlled with a lifelong commitment to blood-sugar monitoring, weight management, nutrition and exercise, making diabetes a treatable but very restrictive condition; however, there is no cure for T2DM (Feld, S. 2002).

Sutherlandia frutescens (*S. frutescens*), a South African traditional medicinal plant, is frequently used to treat a variety of diseases including T2DM (van Wyk & Albrecht, 2008). Although *S. frutescens* has various traditional applications, little scientific evidence is available about its effect on the human body (MacKenzie, Koekemoer, van de Venter, Dealtry & Roux, 2009). For instance, although this plant is traditionally used as an anti-diabetic supplement, no clinical study on its efficacy in a diabetes patient has thus far been conducted. As seen from the rationale developed in Chapter 4, there are solid merits for such a study to be conducted with *S. frutescens* for which this clinical study protocol is required.

Based on the Chapter 4 the sound rationale for testing the efficacy of *S. frutescens* in adult type 2 diabetes patients has been successfully established according to, the information on

literature reported experiences on herbal products clinically tested in diabetes patient and traditional use, chemistry and pre-clinical studies of *S. frutescens*.

Given all the afore-mentioned arguments, the objective of this study was, therefore, to develop a protocol for a trial designed to evaluate the efficacy of *S. frutescens* on several markers of glycaemic control in adult patients with T2DM.

In this chapter, the methods used to design and prepare such a protocol and the major features of the eventually proposed protocol is presented and discussed.

5.2 Methods

To realize the above-mentioned objective the following were achieved.

5.2.1 Review clinical trials of conventional anti-diabetic drugs for type-2 diabetes mellitus

A search was done for clinical trials studies (protocols) of anti-diabetic drugs for the treatment of T2DM by using keywords: Type 2 diabetes mellitus, clinical trial, and efficacy and anti-diabetic drug. The following databases were accessed to collect this information electronically: www.clinicaltrial.gov, Science Direct, PubMed, Google Scholar, journal.pols.com, Cochrane Library Database. Information on the following items features of clinical trials was assessed, viz. study objectives, methods, study design used, type of patients, assessment of the clinical trial information obtained from allopathic drug compared to that typically evident in the clinical trial protocol(s).

5.2.2 Review of existing clinical studies of herbal products in type 2 diabetes mellitus

A medical literature search was first done for all clinical trials in which herbal medicines were used for the treatment of T2DM. Information on the following items (features of clinical trials) was retrieved and assessed, viz. study objectives, methods, study design used, type of patients, type of herbal products tested and assessment of the clinical trial information obtained from the herbal medicine trials was also compared to that typically evident in clinical trial protocol(s). The following databases were accessed to collect this information electronically: www.clinicaltrial.gov, Science Direct, PubMed, Google Scholar, journal.pols.com, Cochrane Library Database, United States of America Food Drug

Administration (USA's FDA), European Medicine Agency (EMA) and South Africa's Medicine Control Council (MCC). In the search activity, the following terms were used, viz. type 2 diabetes mellitus, plant, herb, traditional and natural or herbal medicine. Information on the following items or features of clinical trials was retrieved and assessed, viz. study objectives, methods, study design used, type of patients, type of herbal products tested, and information obtained from the other herbal medicine trials. Overall, this process was expected to identify and confirm all the essential features of a protocol to test *S. frutescens* in T2DM patients.

5.2.3 Preparation of a protocol to test the efficacy of *Sutherlandia frutescens* in adult type 2 diabetes mellitus

To realize the objective of this part of the investigation, the information gained from 5.2.1 was used to establish a protocol for a clinical study to evaluate the efficacy of the herbal preparation – *S. frutescens*. The protocol had to contain all the relevant features, viz: study objective, method, study design, type of patients. Furthermore, the protocol was compiled according to the South African Good Clinical Practice (GCP) guidelines (www.kznhealth.gov.za/research/guideline2.pdf) and had to comply with these. Finally, to be acceptable, we had to assess whether the protocol also complied with the internationally recognized guidelines of the World Health Organization (2012).

5.3 Result and discussion

5.3.1 Review of anti-diabetic drugs for type-2 diabetes mellitus

Searches on the literature for clinical trials on anti-diabetic drugs were done and we present three studies in table 5.1 below with the discussion following thereafter:

Table 5. 1: Three clinical studies of anti-diabetic drugs for type-2 diabetes mellitus

Drug name and references	Study design	Objective	Duration	Dose	Number of patients	Outcome
Pioglitazone Einhorn, Daniel, et al, (2000)	Randomized, Placebo-Controlled Study	To assess the efficacy and tolerability of pioglitazone in combination with metformin in patients with T2DM	16-weeks	Pioglitazone 30 mg + metformin or placebo + metformin, once-daily	328 patients	Decreases in HbA1c, ↓fasting plasma glucose
Canagliflozin Lavalle-González, F. J., et al, (2013)	Randomised, double-blind, placebo- and active controlled, Phase 3 study	To evaluate the efficacy and safety of canagliflozin vs placebo and sitagliptin in patients with T2DM who were being treated with background metformin	April 2010 and August 2012	canagliflozin 100 mg or 300 mg, sitagliptin 100 mg, or placebo	1284 patients	Reduced HbA1c vs placebo Canagliflozin improved glycaemia and reduced body weight vs placebo
Berberine Yin, J., Xing, H., & Ye, J. (2008)	Pilot study	To determine the efficacy and safety of berberine in the treatment of T2DM patients	3-months	Treatment with berberine or metformin (0.5 g 3 times a day)	36 adults with newly diagnosed	Significant decreases in haemoglobin HbA1c, ↓fasting blood glucose, ↓postprandial blood glucose, ↓plasma triglycerides

1. Pioglitazone

(Einhorn, Daniel, et al, 2000) - A 16-week, double-blind study with the option of enrollment in a separate open-ended, open-label study. The study was carried out to assess the efficacy and tolerability of pioglitazone in combination with metformin in patients with T2DM. Included patients were those with poorly controlled diabetes mellitus. Patients were randomized to receive once-daily pioglitazone 30 mg + metformin or placebo + metformin. Patients in the open label extension received pioglitazone 30 mg (with optional titration to 45

mg) + metformin. The results showed that three hundred and twenty-eight (328) patients were randomized to treatment (168 pioglitazone + Metformin, 160 placebo + metformin), and 249 completed the study. Of these, 154 elected to enter the open-label extension study. Patients receiving pioglitazone 30 mg + metformin had a statistically significant mean decreases in HbA1c (-0.83%) and fasting plasma glucose (FPG) levels (-37.7 mg/dL) compared with the placebo + metformin ($P \leq 0.05$) group. Decreases in FPG levels occurred as early as the fourth week of therapy, the pioglitazone + metformin group had significant mean percentage changes in levels of triglycerides (-18.2%) and high-density lipoprotein cholesterol (+8.7%) compared with placebo + metformin ($P \leq 0.05$). Mean percentage increases were noted in low-density lipoprotein cholesterol levels (7.7%, pioglitazone + metformin; 11.9%, placebo + metformin) and total cholesterol (4.1%, pioglitazone + metformin; 1.1%, placebo + metformin), with no significant differences between groups. In the extension study, patients treated with open-label pioglitazone + metformin for 72 weeks had mean changes from baseline of -1.36% in HbA1c, and -63.0 mg/dL in FPG. The incidence of adverse events was similar in both groups (Einhorn, Daniel, et al, 2000).

2. Canagliflozin

(Lavalle-González, F. J, et al, 2013) - A randomised, double-blind, four-arm, parallel-group, Phase 3 study was conducted in April 2010 and August 2012, to evaluate the efficacy and safety of canagliflozin vs placebo and sitagliptin in patients with T2DM who were being treated with background metformin. Participants (N =1,284) primary endpoint was to have a change from baseline in HbA1c at week 26; secondary endpoints included changes in HbA1c (week 52) and fasting plasma glucose (FPG), body weight, and systolic blood pressure (BP; weeks 26 and 52). Adverse events (AEs) were recorded throughout the study. The results reported that, at week 26, canagliflozin 100 mg and 300 mg reduced HbA1c vs placebo (-0.79%, -0.94%, -0.17%, respectively; $p < 0.001$). At week 52, canagliflozin 100 mg and 300 mg demonstrated non-inferiority and canagliflozin 300 mg demonstrated statistical superiority, to sitagliptin in lowering HbA1c. Both canagliflozin doses reduced FPG and systolic BP vs placebo (week 26) and sitagliptin (week 52) ($p < 0.001$). Canagliflozin improved glycaemia and reduced body weight vs placebo (week 26) and sitagliptin (week 52) and was generally well tolerated in patients with T2DM on metformin (Lavalle-González, F. J, et al, 2013).

3. Berberine

(Yin, J., Xing, H., & Ye, J.2008) - In study A, 36 adults with newly diagnosed T2DM were randomly assigned in a 3-month trial to treatment with berberine or metformin (0.5 g 3 times a day), to determine the efficacy and safety of berberine in the treatment of T2DM patients. The hypoglycemic effect of berberine was similar to that of metformin. Significant decreases in hemoglobin A1c (from $9.5\% \pm 0.5\%$ to $7.5\% \pm 0.4\%$, $P < .01$), fasting blood glucose (from 10.6 ± 0.9 mmol/L to 6.9 ± 0.5 mmol/L, $P < .01$), postprandial blood glucose (from 19.8 ± 1.7 to 11.1 ± 0.9 mmol/L, $P < .01$), and plasma triglycerides (from 1.13 ± 0.13 to 0.89 ± 0.03 mmol/L, $P < .05$) were observed in the berberine group. In study B, 48 adults with poorly controlled T2DM were treated with berberine in a 3-month trial. Berberine acted by lowering fasting blood glucose and postprandial blood glucose from 1 week to the end of the trial. Hb A1c decreased from $8.1\% \pm 0.2\%$ to $7.3\% \pm 0.3\%$ ($P < .001$). Fasting plasma insulin and homeostasis model assessment of insulin resistance index were reduced by 28.1% and 44.7% ($P < .001$), respectively. Total cholesterol and low-density lipoprotein cholesterol were decreased significantly as well. During the trial, 20 (34.5%) patients experienced transient gastrointestinal adverse effects (Yin, J., Xing, H., & Ye, J.2008).

5.3.2 Review of existing clinical studies of herbal products in type 2 diabetes mellitus

The extensive search on the literature for clinical trials on herbal medicines lead to more than 30 review articles, out of which we identified specific articles from five studies which complied with this study's criteria of acceptance. These clinical trials were performed in different countries and varied substantially. We included these studies as randomized clinical trials which had been done on herbal treatments in anti-diabetic patients. A summary of these studies are presented in table 5.2 as per criteria with the discussion of the result following thereafter.

Table 5. 2: Clinical studies of herbal medicines for type2 diabetes mellitus

Plant (scientific name) Authors (year)	Study design	Disease/ Effect Target	Duration	Dose	Number of patients	Outcome
<i>Ayurvedic formulation</i> Awasthi <i>et al.</i> (2015)	Randomized control study	T2DM	6 months	polyherbal capsules 500 mg/day +Metformin 500 mg/day	93 patients	↓fasting and post prandial blood glucose, ↓HbA1c, ↓total cholesterol level
<i>Rauwolfia-Citrus</i> Campbell-Tofte <i>et al.</i> (2011)	Randomized and double-blinded pilot study	T2DM	16 weeks	250 mg tea bags	11/15 patients	↓HbA1c, ↓postprandial plasma glucose, ↓fasting plasma glucose
<i>Salacia reticulata</i> Jayawardena <i>et al.</i> (2005)	Randomised single centre double-blind cross over	T2DM	6 weeks	Standard tea bags	28/23 patients	↓HbA1c
<i>Psyllium</i> Ziai <i>et al.</i> (2005)	RCT double-blind	T2DM	8 weeks	10.2 g/day	27/22 patients	↓fasting blood glucose, ↓HbA1c, ↑HDL
<i>Silybum Marianum</i> Huseini <i>et al.</i> (2006)	RCT	TDM2, Hyper-lipidaemia	4 months	600 mg+ Standard therapy	29/25 patients	↓fasting blood glucose, ↓LDL, ↓cholesterol, ↓TG, ↓SGOT, ↓SGPT

All of these studies on herbs and herbal preparations were found to treat diabetic patients, most trials examined herbs or herbal preparations as an adjunct to conventional treatment with diet and/or medication. The most common outcome measures encountered in these studies were fasting- and postprandial blood glucose, hemoglobin A1c (HbA1c), and postprandial plasma glucose and cholesterol levels.

For the five compliant studies identified, the data showed that some of these plants were effective in reducing blood glucose, (Awasthi *et al.*, 2015; Campbell-Tofte *et al.*, 2011; Jayawardena *et al.*, 2005; Huseini *et al.*, 2006). Ziai *et al.*, (2005) stated that blood glucose and HbA1c were significant and increased the cholesterol level, while Huseini *et al.* (2006) showed a significant decrease in HbA1c, fasting blood sugar (FBS), total cholesterol, Low-density lipoprotein (LDL), triglyceride, Serum glutamic oxaloacetic transaminase (SGOT) and serum glutamic-pyruvic transaminase (SGPT) levels in patients treated with *Silybum marianum* compared to placebo in each group. This was also shown in their base-line values at the beginning of the study (Huseini *et al.*, 2006).

In T2DM patients all of the controlled clinical trials suggested the efficacy of these plants. All Randomised Control Trial (RCT) on herbal preparations that were included showed significant effects on T2DM patients. In some of the RCT reviews on *S. frutescens*, Johnson *et al.*, (2007) reported on a study in which healthy adults received 800 mg leaf powder capsules per day. Favourable outcomes and results have been described and indicate that the 800 mg daily dosage was well tolerated by study participants, with no significant changes present in the relevant haematologic, biochemical or physiological parameters (Johnson *et al.*, 2007).

Although the results for some of the trials indicated that anti-diabetic treatment with a herbal medicine could be of possible value, the trial design/methods showed significant imperfections. Therefore, no convincing overall conclusions could be drawn on that point. There were, however, also studies with promising results that warrant further research and analysis.

A review of some studies highlighted the limitations of research into preparations as lack of placebo controlled double blind clinical trials, low sample size, inadequate description of method, lack of statistical analysis and short duration. And assessments into the research method. On the evidence of this trial that has addressed all these limitations.

Overall the brief search suggested that the guidelines for the conduct of clinical trials were not followed and therefore the quality of the clinical trials on herbal medicines was generally inferior to the clinical trials done on regular pharmaceutical products.

Nevertheless, the results of the search for drawing up the protocol on clinical and previous trials on herbal medicines, suggest this could be successfully set up with a focus on the study objective, methodology, informed consent and ethical approval. However, the following concerns need to be correctly addressed: a clinical trial should definitely be randomized and blinded; the duration and outcome of the trial should be scientifically relevant; the dose of the herbal product should be validated and in similar form as the traditionally used product; all information on the historical use of the herbal medicine and any available safety data should be included in the protocol; the inclusion and exclusion criteria must be very precise; and the study population specified and the informed consent should be fully detailed. Furthermore, the statistical analysis must be accurate and fully comply with international standards; and finally, approval by the regulatory and ethical authorities should ensure that the clinical trial is awarded the same prestige as are most clinical trials on regular pharmaceutical products.

5.3.3 A protocol for testing the efficacy of *Sutherlandia frutescens* in adult type 2 diabetes mellitus

To realize the objective of this part of the investigation, the information gained from 5.2.1 and 5.2.2 was used to design a protocol for a clinical study to test the efficacy of the herbal preparation – *S. frutescens* in T2DM. The protocol was designed and contains all the relevant features, viz: study objective, method, study design and type of patients. Furthermore, the protocol was compiled according to the South African GCP guidelines and had to comply with these. Finally, to be acceptable we had to assess whether the protocol also complied with the internationally recognized guidelines as well. For more detail of the full protocol see (Appendix 1) and the synopsis is presented in Table 5.3.

The important features indicated by the various guidelines included in the protocol can be summarized as follows:

- The protocol clearly stated the objective of the clinical trial.
- A detailed plan regarding the methodology, statistical consideration and organization of the trial was given.
- Appropriate provision was made to adequately inform subjects of the aims, methods, sources of funding, potential hazards/discomforts it may entail and the freedom to withdraw consent by means of the patient information leaflet and consent form.

The protocol was designed for a randomized double blind placebo controlled study to investigate the efficacy of *Sutherlandia frutescens* (*S. frutescens*) (sub-species *Microphylla*) in adult type 2 diabetics (T2DM). The objective of this study was to determine the efficacy of *S. frutescens* capsules in adult T2DM. The study design protocol was to do a 6 months study at 1,200 mg/day of *S. frutescens*. Eligible participants were men and women with T2DM, aged ≥ 18 and ≤ 70 years, who had inadequate glycaemic control and hemoglobin A1c (HbA1c $\geq 7.0\%$). The study protocol would be conducted in accordance with ethical principles that comply with the Declaration of Helsinki and be consistent with Good Clinical Practices and applicable regulatory requirements.

All participants would provide written informed consent before taking part in the study.

Table 5. 3: Synopsis of protocol for testing the efficacy of *S. frutescens* on T2DM

TITLE	A randomized double blind placebo controlled study to investigate the efficacy of <i>Sutherlandia frutescens</i> (subspecies <i>Microphylla</i>) in adult type 2 diabetes mellitus.
Investigational site:	Clinical Research Centre, University of Cape Town, L51 Old Main Building, Groote Schuur Hospital, Observatory Road.
Investigators:	TBA
Project Leaders:	Prof. James Syce, Mr. R Swead
Sponsor:	TBA (School of Pharmacy, University of Western Cape)
Clinical Phase:	II
Study number:	UWC 16-002
OBJECTIVES:	<ul style="list-style-type: none"> • To determine the efficacy of <i>Sutherlandia frutescens</i> (subsp. <i>Microphylla</i>) capsules in adult type 2 diabetes mellitus by assessing, HbA1c. • To evaluate the proportion of participants achieving a therapeutic response (HbA1c <7%) with <i>S. frutescens</i> 1,200 mg versus placebo after 12 weeks of treatment. • To determine the effect of <i>S. frutescens</i> on glycaemic control (HbA1c, fasting plasma glucose, etc.).
STUDY DESIGN:	A 6-month randomized double-blind placebo-controlled study. Participants will be screened at Visit 1 and eligible subjects will, at visit 2, be randomized to either <i>S. frutescens</i> (subsp. <i>Microphylla</i>) or placebo. 1,200 mg capsule, b.i.d or a placebo with a similar appearance.
Participants:	<p>Inclusion criteria</p> <ol style="list-style-type: none"> 1. T2DM men and women between 18–70 years of age on a stable medication. 2. HBA1C >6.5% <8.5% 3. Fasting plasma glucose (FPG) 7.0–14.0 mmol/l 4. Have body weights within 25% of the appropriate weight range 5. Had taken standardized diet control + exercise therapy + stable administration of metformin for more than 3 months before being enrolled. 6. Has a normal ECG as determined by a unit physician. 7. Has not taken any traditional medication for 28 days prior to screening. 8. Be informed of the nature of the study and will give written informed consent. <p>Exclusion criteria</p> <ol style="list-style-type: none"> 1. Type 1 diabetes mellitus, gestational diabetes mellitus and other special types of diabetes mellitus. 2. In the past 3 months, had taken medication to control body weight (including weight-loss drug) or oral administration of any anti-diabetic drugs or insulin except for metformin. 3. Had diabetic ketosis, ketoacidosis. 4. Fasting triglycerides >450 mg/dL (>5.1 mmol/L) 5. Had uncontrolled hypertension (blood pressure be and more than 160/100 mmHg). 6. Intake of anti-diabetic medication within 14 days before the start of the study. 7. Had mental illness, alcohol addiction and /or taken administration of psychoactive drug substance. 8. Smokers, who smoke more than 10 cigarettes per day and cannot, refrain from smoking during the study period. 9. Pregnancy, lactation or being prepared, pregnant woman 10. Had allergic constitution. 11. Unwillingness or inability to follow the procedures outlined in the protocol. 12. Any disease or condition which might compromise the haematopoietic, renal, endocrine, pulmonary, central nervous, cardiovascular systems.
PRODUCT TO BE EVALUATED	<p>Test: <i>Sutherlandia frutescens</i> 1,200 mg/day, capsules.</p> <p>Reference: Placebo capsules</p>
DURATION OF STUDY	6 months ±30 days

ENDPOINTS:	<p>Primary Endpoint: primary endpoints are defined as weight loss and change in HbA1c levels comparing between the treatment groups.</p> <ol style="list-style-type: none"> 1. Vital parameters: pulse rate, blood pressure, respiratory rate, weight, and height will be recorded at the baseline visit only. 2. Adverse events and serious adverse events. To follow a non- exhaustive list and any adverse event that patients reported will be tracked. These events are as follows: <ol style="list-style-type: none"> a) CNS (e.g. headaches, nervousness, dizziness) b) GIT (e.g. diarrhoea, gastritis, constipation, stomach cramps, nausea). c) Infections d) Allergy (e.g. dermatitis). e) Appetite (e.g. increase, decrease). f) General (e.g. iliac pain). g) Others <p>The following primary efficacy precautions will be measured per the flowchart on the page.</p> <ol style="list-style-type: none"> 1. Full-count blood and differential count. 2. Physical examination. 3. 12-lead electrocardiogram. <p>Secondary Endpoints: The following endpoints will be measured at the baseline, week 4, week 12, and week 24;</p> <p>Efficacy Endpoints: The following efficacy endpoints will be obtained at the designated study visits:</p> <ol style="list-style-type: none"> 1. Medical history. 2. Medical symptoms. 3. Alcohol/substance use. 4. Patient Satisfaction with the study.
SAMPLE SIZE:	100 subjects
STATISTICAL ANALYSIS:	<p>An intention to the treatment approach was used. The primary endpoint for efficacy studies would be the end result of treatment with either the placebo or the drug. The data would be analyzed as a within patient comparison of treatments using the t-test for paired differences. The t-test would also be used to test for to the period effect and the carry over effect.</p>

Official Title: A randomized double blind placebo controlled study to investigate the efficacy of *Sutherlandia frutescens*_(subspecies *Microphylla*) in adult type 2 diabetics

Investigational site: Clinical Research Centre, University of Cape Town, L51 Old Main Building, Groote Schuur Hospital, Observatory Road

Project Leaders: Prof J Syce, Ramadan Swead

Investigators: N/A

Sponsor: N/A

Study Design: Allocation: Randomized

Masking: Double-Blind

Purpose of the study: The aim of this study is to investigate the efficacy of *Sutherlandia frutescens*_(subsp. *Microphylla*) capsules in adult type 2 diabetes mellitus by assessing, HbA1c

Eligibility:

Ages eligible for study: 18 Years and older

Sexes eligible for study: All

Accepts healthy volunteers: No

PRODUCT TO BE EVALUATED: Test: *Sutherlandia frutescens* 1,200 mg/day, capsules.

Reference: Placebo capsules

Enrollment: 100

DURATION OF STUDY: 6 months

The inclusion and exclusion criteria and primary endpoint and secondary endpoint will be follow as set up clearly in the protocol for further details see appendix 1 full protocol.

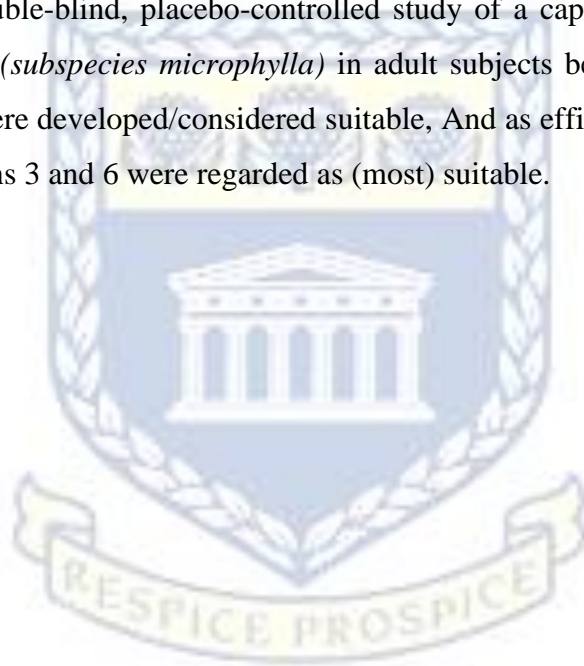
Acceptability of the protocol:

To test the protocol acceptability: The protocol will be submitted to the regulatory and ethics board for approval i.e., to the MCC and ethics committee (university of Stellenbosch),

respectively. In addition the comments from two experts will be solicited. It will be assumed that approval of the protocol by either the regulatory or ethics authority would be the major measure of the acceptability of the study.

5.4 Conclusion

A protocol was designed according to the South African GCP guidelines and is ready to be carried out to assess the efficacy of the traditional plant *S. frutescens* for the treatment of adult patients with T2MD. To test for the efficacy of *S. frutescens* in diabetes a protocol for a 6 month, randomized, double-blind, placebo-controlled study of a capsule form of 1,200 mg dosage of *S. frutescens* (*subspecies microphylla*) in adult subjects between ages 18 and 65 years, having T2DM, were developed/considered suitable, And as efficacy parameter HBA1c measured twice at months 3 and 6 were regarded as (most) suitable.



UNIVERSITY of the
WESTERN CAPE

Chapter 6

Application documentation required for regulatory approval of an efficacy trial of *Sutherlandia frutescens* in adult type 2 diabetes mellitus

6.1 Introduction

In South Africa, the Medicine Control Council (MCC), as the regulatory authority, has the responsibility for ensuring that all clinical trials of non-registered medicines and new indications of registered medicines comply with the necessary requirements for efficacy and safety and proof of safety, quality and efficacy must be submitted when applying to the MCC for approval and registration of a medicine for use in South Africa. (Ngcobo *et al.*, 2012). Specific regulations for the registration and control of new traditional herbal medicines have not yet been fully established and implemented in South Africa (Ngcobo, Nkala, Moodley, & Gqaleni, 2012), but a few herbal medicines, such as *Senna* and *Aloes*, have internationally been tested for their efficacy and safety and registered (Calixto, 2000 & Pradesh, A., 2011). The MCC is expected to oversee the registration and regulation of the practice of traditional medicine (TM) by setting practice standards and applications for clinical trials and for registration of medicines and medical devices are reviewed by an MCC expert committee which considers, amongst other issues, the scientific, medical and ethical issues of the applications. Reports on the progress of the approved studies already implemented are also sent to the MCC on a regular basis.

Sutherlandia frutescens (*S. frutescens*) is a South African traditional medicinal plant that is used to treat a variety of diseases, including type 2 diabetes mellitus (T2DM) (Faleschini, Myer, Harding & Fouche, 2013). However, thus far, there is no record of any regulatory approved efficacy trial having been conducted on *S. frutescens* in the treatment of T2DM in South Africa or elsewhere in the world.

Given all the afore-mentioned arguments, two objectives of this project were therefor to (i) establish the regulatory requirements that need to be met for such a clinical trial and (ii) to prepare the specific documentation that would be needed to obtain regulatory approval from the certified South African medicine regulatory authority, viz. the MCC, for a trial to establish the efficacy of *S. frutescens* in adults with T2DM.

In this chapter, the regulatory requirements for such a trial are reviewed and the application documentation for such regulatory approval prepared and discussed.

6.2 Methods

To realize the above-mentioned objectives, the following methodology was applied.

6.2.1 Determination of the general regulatory requirements for traditional herbal medicines

An Internet search was performed for information on the regulatory requirements for traditional herbal products in South Africa and internationally. The data retrieved was examined and assessed for criteria which could be used as requirements for approval and registration of traditional herbal medicines. In the search, the following keywords and phrases were used: “regulation of herbal products”, “herbal medicines”, “guidelines of herbal medicines”, and “requirements for registration of herbal medicines”. The websites that were searched included those of the European Medicine Agency (EMA), World Health Organisation (WHO), Google Scholar and PubMed. The search was conducted in April and June 2016 and from the information gathered a list of regulatory requirements that might be appropriate for approval of herbal medicines was drawn up.

6.2.2 Preparation of an application for regulatory approval for testing *Sutherlandia frutescens* in type 2 diabetes mellitus

First, the guidelines and specific requirements for an application for regulatory approval for the *Sutherlandia* trial by the MCC were obtained from this regulatory authority’s website at www.mccza.com. Then all the necessary forms for the MCC application were completed, and a special note made regarding the requirements of the MCC and those of various other regulatory authorities to see how these requirements agreed or disagreed. The forms and instructions available to researchers seeking regulatory approval from the MCC were used and completed. After completion of the application, two experienced clinical investigators and one clinical trial coordinators reviewed the documentation submitted. In addition independent clinical trial monitors were also be asked to review and advise on the appropriateness of the final regulatory application.

6.3 Results and discussion

6.3.1 The regulatory requirements for traditional herbal medicine

A summary of the regulatory requirements of various regulators for registration of herbal medicine is presented in table 6.1 and discussed below

Table 6. 1: A summary of regulatory requirements for the registration of traditional herbal medicines*

Requirements for registration of traditional medicine (TM)	Food and Drug Administration (United States of America) ^a	European Medicine Agency (European Union) ^b	Australian Guideline for Complementary Medicines ^c	WHO Guidelines for Registration of Traditional Medicine ^d	Medicine council control (South Africa)
Traditional use	Not considered	Traditional use does not substitute quality. Bibliographical evidence of traditional use must be presented	Use of TM must be well established and widely acknowledged; effective preparation, dosage, method of use and indications are well established; botanical identification of herbal substances	Traditional use not addressed directly but it is considered in evaluation of safety and efficacy of TM	The use of such medicines must be well established between many THPs over a long time and widely acknowledged. The efficacy must be credible on the basis of long-term use and experience. The botanical identity of the plant must be known and proven through bibliographical evidence.
Pharmaceutical quality	As conventional drugs	Starting materials must be defined by botanical identification and quality must be adhered to until the final herbal product	Each ingredient in formulation should be characterized; state the plant part used and processing before use in manufacture of the product; brief description of manufacturing process must be provided	Raw plant material(s) should be identified; state plant part used; chemical, physical & biological tests for general identification; finished product to be qualitatively and quantitatively analyzed	Ensuring quality of TM products is a shared responsibility of the MCC and the manufacturers of the products, must be enforced to ensure pharmacovigilance in collection of raw materials and manufacturing of the finished products.
Stability testing	As conventional drugs	Stability should be determined by appropriate fingerprint Chromatograms	Stability data must be sufficient to demonstrate that the product intended for market will remain safe, of consistent quality and efficacious throughout its shelf life. Maximum permitted shelf life is 5 years	Stability tests fall under quality evaluation of raw and finished product	Should be determined by appropriate chromatographic fingerprinting and other specific sensory and physical tests.
Safety	As conventional drugs	Bibliographical evidence of safety including cross-referencing to expert report. Safety summary including herbal ingredients, use in pregnancy & lactation and possible drug interactions must be included	Traditional use not a substitute for safety. Long-term and safe therapeutic use will be considered in evaluating the safety of products. Safety summary including herbal ingredient uses in pregnancy and lactation and possible drug interactions must be included	Botanical authentication of raw plant(s); biological information obtained via literature/database search; standard toxicological studies mandatory even of long-term use history to be available	Should not be a substitute for safety but must be taken into consideration when safety is evaluated. Where traditional use is used as safety evidence, the details of use (duration, route, dose etc.) should be consistent with the proposed use.
Efficacy	As conventional drugs	Efficacy must be plausible on the long-standing use and experience	Traditional use will be taken into account when establishing efficacy. Where traditional use is not sufficient, clinical evidence data should be provided.	Efficacy is dependent on kind of indications for use & individual experiences of doctors, traditional health practitioners or patients. Clinical evidence is required for traditionally-used plants in case of a new indication. Active ingredients should be standardized if known; if not known, the TM as a whole should be regarded as ONE active ingredient.	Long-term use of TM will have to be considered when efficacy is evaluated. Bibliographical evidence from herbal books, pharmacopeias, peer-reviewed scientific papers etc., must be presented along with the application to prove traditional use. The type of evidence presented will depend on the kind of indications for use and individual experiences as recorded in reports from physicians, THPs or treated patients.
Labelling and packaging	As conventional drugs	Packaging leaflet shall reflect the results of consultations with target patient groups to ensure that it is legible, clear and easy to use	Labels should reflect all the necessary information about the product as guided by the <i>Australian Therapeutics Goods Act 1989</i>	Labelling and packaging is addressed under quality control and good manufacturing practices of the finished product	Labels reflect the results of consultations with target patient group groups to ensure that it is legible, clear and easy to use. Packaging should form part of the GMP requirements for manufacturers of TM products.
Multi-ingredient products	Each product must be clearly identified and tested separately	Quantitative determination of each active substance separately	Traditional formulations will normally be accepted unless current adverse evidence exists. New formulations should provide information of efficacy of each herbal ingredient	Multi-ingredient products not mentioned directly but the guidelines do mention formulations of herbal medicines in quality, safety and efficacy assessment	Multi-herbal product should have each plant tested separately for quality, safety, efficacy and stability.

* Ngcobo *et al.*, 2012 (a FDA, 2006; b EMEA, 2006; Deal, 2010; c Australian government, 2005; d WHO, 2004)

(In essence the regulators may have up to 6 issues they might consider in their requirements for the registration of a traditional medicine. These are/include, such as Traditional use, Pharmaceutical quality, Stability testing, Safety, Efficacy)

Basically the USA FDA has developed its own draft guidelines for the regulation of complementary and alternative medicines (CAM). It has devised four domains to classify and regulate CAMs, and TMs might fall into one or two of these domains; either as biologically-based practices or whole medical systems. Botanical products, which include the traditional herbal medicines, typically fall under the biologically-based product domain. Any new herbal product identified as treatment for a specific disease is therefore treated as any new conventional drug and will have to meet similar safety and efficacy requirements (FDA, 2006). The European Medicines Agency has similar registration requirements but also states that the quality of a medicinal product is independent of its traditional use and that all general principles of quality therefor also apply to traditional herbal medicines intended for human use (EMA, 2006a, 2006b; Deal, 2010).

The Australian regulatory guidelines have references to the traditional use of herbs or herbal mixtures and to multi-ingredient products. According to these guidelines, traditional use is not a substitute for safety but will be considered in the evaluation of the efficacy of the herbal medicine. Traditional multi-ingredient products are normally accepted unless current adverse evidence exists. New formulations are assessed as any other new drug (Australian government, 2005a). The WHO guidelines for registration of TM in the African region provide the Member States with the framework for the development of regulations. These guidelines provide the minimum requirements for registration of TM regarding pharmaceutical quality, safety, stability testing, therapeutic efficacy, and labelling and packaging. Because of the diversity of the type of TM product in Africa, the WHO guidelines have created four TM categories to simplify the implementation of mechanisms of registration. These categories range from traditional health practitioners (THP) remedies, prepared for individual patients, to TM derived from scientific research (WHO, 2004). Therefore, South Africa, as a Member State of the WHO, has the necessary framework to develop regulations for the registration of TM.

The regulations and guidelines for registration of TM are aimed at all stakeholders, including national drug regulatory authorities; in the case of South Africa these include the MCC, THP, manufacturers and drug research institutions. Traditional health practitioners and the TM system of the country, therefore, need to be developed along with the development and

implementation of regulations for registration of TM. This can be achieved by regulating the THP practices and having all THP registered. This will allow THP to be part of the processes of development of regulations that will control their medicinal products. This will serve as a great educational tool for the THP involved in this process and they can in turn be deployed to educate other healers. The use of pharmacopeias and monographs for the naming of TM products will require a good understanding from THP, who will want to have their products registered. The idea of involving THP in all these processes is simply because they are the knowledge holders and their involvement will help to safeguard their intellectual property.

Efficacy as a regulatory requirement:

In table 6.1, we highlight ‘efficacy’ as a regulatory requirement for the registration of TMs since the study title and focus is the ‘preparation of the documentation required for regulatory approval of an efficacy trial of *Sutherlandia frutescens* in adult type 2 diabetes mellitus. A worthy argument has been made for the different regulatory bodies for the approval to test the efficacy of TM. The US (FDA) has classified the registration of their TMs as per ‘conventional drugs’ and will thus have to meet similar requirements. It is believed that TMs is safe for human use based on the THPs knowledge and experiences working with them and their patients, and often toxicity being reported are associated with miss-identified medicinal plant species. The scientific validation is necessary to understand safety for those undocumented medicinal plants as to safe-guide consumers with the issues of poisoning by unknown medicinal plants (Pillay et al., 2011; Fennel et al., 2004). New evidence might be required for new formulations of TMs. This new evidence should address issues related to pharmacodynamics, pharmacokinetics, bioavailability and possible clinical trials. We recommended that it would be more feasible for THPs to have their TM products evaluated by *in vitro* and *in vivo* animal models for their pharmacodynamics, pharmacokinetics, and bioavailability effects. Pharmacodynamics details the possible mechanism of action, dose and dose intervals, and possible drug interactions. Pharmacokinetics describes what happens inside the body once the medicine is taken by the patient and includes the absorptions, distribution, metabolism and elimination. Bioavailability describes the proportion of administered medicine reaching systemic circulation. (Australian government, 2005b). With effective ‘efficacy’ evidence, *S. frutescens* will thus prove to be safe in both healthy and T2DM volunteers which will enable its registration by the SA MCC.

6.3.2 Application for regulatory approval for testing *Sutherlandia frutescens* in type 2 diabetes mellitus

The MCC of South Africa is committed to ensuring that all registered medicines will be of the required quality, safety, and efficacy. Therefore, it is important that applicants adhere to the administrative requirements to avoid delays in the processing and evaluation of applications. On the website, www.mccza.com, all the necessary information and forms that had to be completed to apply for approval to this committee were downloaded. These documents and information were found under the tab, 'Forms' on the afore-mentioned web address. For submitting the clinical trial application, information on twenty-one items were required as follows:

1. Covering letter.
2. Cover sheet.
3. Checklist.
4. Completed application form.
5. All documents to be submitted in duplicate with two electronic copies.
6. Additional 25 copies of the application form must also be submitted.
7. Protocol.
8. Patient Information leaflet and Informed consent form.
9. Standardized MCC contact details/wording to be added to PILs.
10. Investigators Brochure/Package inserts.
11. Signed CV(s) of the investigator(s) in the format required by the MCC.
12. Signed declaration by principal investigator(s).
13. Signed joint declaration by sponsor/national principal investigator.
14. Signed provisional declaration by co- or sub-investigators.
15. Signed declaration by regional monitor.
16. Indemnity and Insurance Certificate and/or.
17. Proof of malpractice insurance certificate of trialist(s).
18. Ethics committee(s) approval or.
19. A copy of the letter submitted to Ethics Committee(s).
20. Diskettes to be submitted in word format.
21. Financial declaration.

The complete application eventually prepared included all the information as requested. These documents are attached as Appendix 2. All the literature available on the historical use and data safety of the plant is included. The application is thus ready for submission for ethics committee. After submission, it is expected that the MCC reviews will focus on the safety data of the plant which is presented. Overall, it is expected that the MCC accepts and approves the application for this study to be conducted.

6.4 Conclusions

The use of herbal drug products is very significant, and a fairly good agreement among the various regulatory agencies regarding the requirements for registration of herbal medicine was noted. An application for approval from the South African medicines regulator (i.e. MCC) that should meet all of the agency's requirements could readily/easily be prepared for this trial. The fact that at least some safety data for *S. frutescens* is available and that a fairly standard diabetes clinical trial design are proposed should facilitate the approval of this trial by the MCC (can be considered as a particular strong point of such an application). In addition the fact that the product(pharmaceutical) quality of dosage form (i.e. capsule) that is to be used can relatively easily be ensured is/should be another a strong point of the application.

Chapter 7

Application documentation for ethics approval for a clinical trial of *Sutherlandia frutescens* in type 2 diabetes mellitus

7.1 Introduction

In South Africa, the Research Ethics Committee (REC) has the responsibility of ensuring the protection of the rights, safety and well-being of all potential participants involved in a clinical trial. The REC provides public assurance of that protection by, among other things, reviewing and approving or rejecting the study protocol and ensuring that the investigator(s) are suitably qualified to conduct the trial. The facilities have to be adequate, and the methods and materials to be used in obtaining and documenting informed consent of the trial participants have to be appropriate. In the execution of these responsibilities, committees should be guided by the relevant current South African ethical guidelines. (Karlberg & Speers, 2010; Department of Health, 2006).

For this thesis a clinical trial to determine the efficacy of *S. frutescens* in T2DM were developed and for such a trial ethical approval would be required. Consequently, the objectives of this part of the thesis was to (i) review the ethical requirements that need to be met for such a clinical trial, and (ii) to prepare the specific documentation that would be needed to obtain ethical approval from a certified South African REC for such a clinical trial of a herbal product.

In this chapter, the ethics requirements for such a trial were reviewed and listed and the application documentation for ethical approval prepared and assessed.

7.2 Methods

To realize the above-mentioned objectives, the following were done.

7.2.1 Determination of the ethics requirements for an efficacy trial of *Sutherlandia frutescens* in type 2 diabetes mellitus to be conducted in South Africa

First, the Internet was used to search for information on the typical ethics requirements for testing herbal products in human subjects and this information assessed for possible requirements and criteria that must be met to obtain an ethically approved clinical trial of *S frutescens*. The following keywords were used in the search: “ethics of herbal products”, “herbal medicines”, “guidelines of herbal medicines” and “requirements for registration of herbal medicines”. The websites searched included: , Google Scholar, PubMed, Science Direct, Google and the website of the Research Committee(s) of the University of Stellenbosch (<https://www.sun.ac.za/english/search/Pages/results.aspx?k=ethics>). This internet search was done in the time period “April 2016” to “July 2016 year”. From the information gathered a list of ethical requirements that may be appropriate to obtain registration and ethical approval of a clinical trial of a *S. frutescens* capsule dosage form was drawn up.

7.2.2 Preparation of an application for ethics approval to test *Sutherlandia frutescens* in type 2 diabetes mellitus

For this part of the study it was decided to focus on preparing the application for submission to a certified local ethics committee, viz the Health Ethics Research Committee of Stellenbosch University. The Stellenbosch University’s health research ethics committee (HREC) functions in accordance with the South African Good Clinical Practice (GCP) guidelines and is recognized by the medicine control council (MCC) of South Africa as one of the committees in the Western Cape that can provide ethical approval for randomized controlled trials. In the period “31 August 2016 an internet search was done and the specific requirements for the application to this ethical committee located at the Tygerberg campus (Cape Town) were obtained via website address, (<https://www.sun.ac.za/english/search/Pages/results.aspx?k=ethics>). Listed and assessed, and then used to prepare a test application document that may be used to obtain ethical approval for the *S. frutescens* trial.

7.3 Results and discussion

7.3.1 Review of the ethics requirements an efficacy trial of *Sutherlandia frutescens* in type 2 diabetes mellitus

Many ethical guidelines and international statements have been produced to facilitate the ethical conduct of clinical trials. These include those formulated in or by the Declaration of Helsinki (1964), WHO, Council for International Organizations of Medical Sciences (CIOMS) Ethical Guidelines (2002), International Council for Harmonisation ICH-GCP – Guidance on Good Clinical Practice (2002), the United Nations Educational Scientific and Cultural Organization (UNESCO), Universal Declaration on the Human Genome and Human Rights (1997), the Council of Europe Convention on Human Rights and Biomedicine (Oviedo Convention, 1997), and its Additional Protocol on Biomedical Research (2005) as well as the European Directive 2001/20/CE. The main objective of these guidelines and international statements are to regulate the conducting of clinical research and facilitate ethical practice in clinical trials. Many of these documents were written in response to specific events and to avoid future scandals. By focusing on the instigating issues, these guidelines tend to emphasize certain ethical requirements while eliding others. Globally, ethical requirements for clinical research aim to minimize the possibility of exploitation. Research subjects are thereby guaranteed treatment with respect, while they contribute to social benefits. Moreover, the main ethical requirements needing careful contemplation are the protection of participants in clinical research (Altavilla, 2013). According to international ethical guidelines, ethically sound research must satisfy a number of important procedural requirements, including choosing the appropriate research endpoints and design; selecting participants equitably; ensuring that risks are reasonable in relation to potential benefits, obtaining voluntary informed consent in writing; providing for adequate care of and compensation to participants for injuries directly sustained during research (subscribing insurance contracts); guaranteeing the confidentiality of personal data; providing appropriate treatment to participants during and after the trial; and obtaining prior review of research by an institutional review board or independent ethics committee (IRB/IEC). Finally, each individual, involved in conducting a trial, should be qualified by education and training (Emanuel, Wendler & Grady, 2000; Altavilla, 2013).

In essence there were 7 requirements that provided a systematic and coherent framework for determination of ethics approval for clinical research (Table 7.1). These ethical requirements are listed in chronological order from the conception of the research to its formulation and implementation. They are meant to guide the ethical development, implementation and review

of individual clinical protocol. Moreover, the 7 requirements are intended to elucidate the ethical standards specific for clinical research and assume general ethical obligation such as intellectual honesty and responsibility (Emanuel, & Grady, 2000). In addition, these ethical requirements are universal and comprehensive but must be adapted to the particular social context in which the research is implemented. The seven requirements are reviewed by definition in table 7.1.

Table 7. 1: Seven requirements for determining whether a research trial is ethical

Ethical requirements	Definition
Social value	Knowledge gained from the research should have the potential to lead to new generalizable knowledge or improvements in health. Partners should specify in advance to whom benefits will accrue and in what way.
Scientific validity	Research should be designed to produce beneficial and generalizable knowledge. This includes designing research so that it can be feasibly implemented in the settings where it will be conducted.
Fair subject selection	Clinical research must not enrol preferentially vulnerable patients into risky studies or privileged patients into studies with a high likelihood of benefits. Enrolment should aim primarily to achieve the scientific objectives of the research and secondarily to minimize risks, to enhance value, and to be conducted efficiently. Subjects should be selected on the basis of scientific importance, not based on convenience, vulnerability or bias.
Favourable risk-benefit ratio	Clinical research should be designed to minimize risks and ensure that the risks are proportional to the potential benefits of the individual subjects and the expected knowledge gained for society.
Independent review	Independent review ensures that clinical research fulfils ethical requirements to protect subjects and ensure public accountability.
Informed consent	Information relevant to the clinical research must be disclosed to subjects so they can understand what it involves and voluntarily give consent to participate in the research.
Respect for enrolled subjects	Clinical research must respect subjects by: <ul style="list-style-type: none"> (1) permitting withdrawal; (2) protecting privacy through confidentiality; (3) informing subjects of newly discovered risks or benefits; (4) informing subjects of the results of the research; and (5) protecting subject welfare.

*Adapted from Emanuel *et al.*2001.

Any application for the ethical approval of the clinical efficacy trial of *Sutherlandia frutescens* in type 2 diabetes mellitus thus had to fulfil these 7 requirements.

7.3.2 Application for ethics approval for testing efficacy of *Sutherlandia frutescens* in type 2 diabetes mellitus

All medical research involving human participants must undergo an independent ethical review. The REC which undertakes the review must be accredited by the national health REC. In the evaluation of clinical trial protocols or study application, the REC must ensure that participants are protected in accordance with international standards and guidelines. For the clinical trial of *Sutherlandia frutescens* in type 2 diabetes mellitus it was decided to use the Health Ethics Research Committee (HREC) of Stellenbosch University to obtain ethics approval for the trial. Details of the specific information required by this REC is presented in Appendix 3A. The specific information required by the chosen REC and how to present it were quite clear and specific, and fully reflected that needed to assess whether the trial would be ethical (i.e. see addressed the requirements identified in Table 7.1).

Using the guidance and information requested in the Appendix 3A a full application for ethics approval for the *Sutherlandia frutescens* efficacy protocol in T2DM was completed. The full application is presented in Appendix 3B. The application must now just be submitted to the HREC to obtain their approval and or additional requirements.

7.4 Conclusions

The results of this part of the study, firstly, provided a clear list of ethics requirements that needed to be met to obtain ethical approval for an efficacy trial of a capsule product of *Sutherlandia frutescens* in adults with type 2 diabetes mellitus. Secondly, all the information and documentation required and requested by a fully certified South African REC could easily be supplied in a standard application for ethics approval. Finally, although the application has not yet been submitted to the REC it is however expected that it adequately addressed and complied with most/all of the typical ethics requirements (e.g. safety of subjects, appropriate design of trial, number of subjects exposed to medication, patient consent and confidentiality issues,) that needed to be considered by the REC.

Chapter 8

Conclusions and recommendations

The overall aim of the present project was to develop an ethically and regulatory approved clinical protocol for the evaluation of the efficacy of *Sutherlandia frutescens* (*S. frutescens*) in type-2 diabetes mellitus (T2DM). The specific objectives of this project were to, (i) establish a scientifically sound rationale for such a clinical study to evaluate the efficacy of *S. frutescens* in adult T2DM; and (ii) design and develop a clinical trial protocol; (iii) prepare an effective application document for the regulatory approval and (iv) prepare an effective application document for ethics approval of such a study.

From the results obtained in this study the following conclusions could be drawn:

1. Based on information on the traditional use, chemistry and results of existing pre-clinical and clinical safety studies of *S. frutescens*, as well as literature reported experiences on similar herbal products clinically tested in diabetes, a sound rationale for testing the efficacy of *S. frutescens* in adult type 2 diabetes patients was successfully established.
2. To test for the efficacy of *S. frutescens* in diabetes a protocol for a 6 month, randomized, double-blind, placebo-controlled study of a capsule form of 1,200 mg dosage of *S. frutescens* (*subspecies microphylla*) in adult subjects between ages 18 and 65 years, having T2DM, was developed/considered suitable, And as efficacy parameter HBA1c measured twice at months 3 and 6 were regarded as (most) suitable.
3. An application for approval from the South African medicines regulator (i.e. MCC) that should meet all of the agency's requirements could readily/easily be prepared for this trial. The fact that at least some safety data for *S. frutescens* was available and that a fairly standard diabetes clinical trial design are proposed should facilitate the approval of this trial by the MCC (can be considered as a particular strong point of such an application). In addition the fact that the product(pharmaceutical) quality of dosage form

(i.e. capsule) that is to be used can relatively easily be ensured is/should be another a strong point of the application.

4. A standard application for ethics approval of the recommended trial was easily prepared. It (should be able to) comply with the most/all of the typical ethics requirements e.g. safety of subjects, appropriate design of trial, number of subjects exposed to medication, patient consent and confidentiality issues, could all adequately be addressed. Could be prepared to access the efficacy of *S. frutescens* in adult type2 diabetes mellitus.

Collectively, the above conclusions lead to the recommendation that:

- 1) The trial test products (active and placebo) be made and their pharmaceutical quality be tested established and documented. In same way as in the guidelines on GCP i.e as any substance or combination of substances which has a therapeutic, prophylactic or diagnostic proupse, or is intended to modify phsiological function, and is presented in a dosage form sutible for administration to human in a such tiral study.
- 2) The MCC & ethics approval applications be submitted for consideration to accept and approve the protocol of this clinical study for this plant (*S. frutescens*) and register the plant for such condition to treat T2DM in South Africa.
- 3) The protocol be submitted for costing. Costs associated with the implementation of clinical trials have become an increasingly important issue, yet little has been done to develop cost reduction approaches and organize efforts to improve clinical study efficiency and performance.
- 4) Implementation of the trial be considered according to the approval from the committees, and furthermore take this study more to test and confirm its efficacy in such disease.

References

- Aalto, A. M., Uutela, A., & Aro, A. R. (1997). Health related quality of life among insulin-dependent diabetics: disease-related and psychosocial correlates. *Patient Education and Counseling*, 30, 215-225.
- Abramson, B. (2005). Nitric oxide a novel mediator of inflammation, NYU Medical Center, Department of Medicine (Rrheumatology) and Pathology and hospital for joint diseases and administration, [<http://www.med.nyu.edu/Research/S.Abramson-res.html>] accessed: 11.06.16.
- Ahmad, I., Aqil, F., & Owais, M. (Eds.). (2006). *Modern phytomedicine: Turning medicinal plants into drugs*. Publisher John Wiley & Sons.
- Altavilla, A. (2013). Ethical Standards for Clinical Trials. in *Legal and Forensic Medicine* (pp. 1117-1126). Publisher Berlin Heidelberg: Springer.
- American Diabetes Association. (2009). Diagnosis and Classification of Diabetes Mellitus. *Diabetes Care*, 32, S62-S67. doi:10.2337/dc09-S062
- Atlas, D. (2006). International diabetes federation. Press Release, Cape Town, South Africa, 4.
- Australian Government, Department of Health and Ageing, Therapeutic Goods Administration, (2005a). Australian regulatory guidelines for complementary medicines (ARGCM) part IV: general guidance.
- Australian Government, Department of Health and Ageing, Therapeutic Goods Administration, (2005b). Australian regulatory guidelines for complementary medicines (ARGCM) part I: registration of complementary medicines.
- Avula, B., Wang, Y. H., Smillie, T. J., Fu, X., Li, X. C., Mabusela, W., & Khan, I. A. (2010). Quantitative determination of flavonoids and cycloartanol glycosides from aerial parts of *Sutherlandia frutescens* (L.) R. BR. by using LC-UV/ELSD methods and confirmation by using LC-MS method. *Journal of Pharmaceutical and Biomedical Analysis*, 52(2), 173-180.
- Awasthi, H., Nath, R., Usman, K., Mani, D., Khattri, S., Nischal, A., & Sawlani, K. K. (2015). Effects of a standardized *Ayurvedic* formulation on diabetes control in newly diagnosed Type 2 diabetics; a randomized active-controlled clinical study. *Complementary Therapies in Medicine*, 23(4), 555-561.
- Bansal, D. (2010). Research methodological issues in evaluating herbal interventions. *Medicine (Baltimore)* 10, 11.
- Bates, S. H., Jones, R. B., Bailey, C. J. (2000). Insulin-like effect of pinitol. *British Journal of Pharmacology*, 130, 1944-1948.
- Bent, S. (2008). Herbal Medicine in the United States: Review of Efficacy, Safety, and Regulation: Grand Rounds at University of California, San Francisco Medical Center. *Journal of General Internal Medicine*, 23, 854-859. doi:10.1007/s11606-008-0632-y

Bethke, P. C., Bodger, M. R., & Jones, R. L. (2004) Apoplastic synthesis of nitric oxide by plant tissue. *The Plant Cell*, 16: 332-341.

Black, N. (1996). Why we need observational studies to evaluate the effectiveness of health care. *British Medical Journal*, 312(7040), 1215.

Boulton, A. J., Vinik, A. I., Arezzo, J. C., Bril, V., Feldman, E. L., Freeman, R., Malik, R. A., Maser, R. E., Sosenko, J. M., & Ziegler, D. (2005). Diabetic neuropathies: a statement by the American Diabetes Association. *Diabetes Care*, 28, 956-962.

Brown, L., Heyneke, O., Brown, D., Van Wyk, J. P. H., & Hamman, J. H. (2008). Impact of traditional medicinal plant extracts on antiretroviral drug absorption. *Journal of Ethnopharmacology*, 119(3), 588-592.

Buyschaert, M., Dramaix, A. S., Wallemacq, P., & Hermans, M. (2000). Hyperhomocysteinemia in type 2 diabetes: Relationship to macro angiopathy, nephropathy and insulin resistance. *Diabetes Care*, 23, 1816-1822.

Cade, W.T. (2008). Diabetes-Related Microvascular and Macrovascular Diseases in the Physical Therapy Setting. *Physical Therapy*, 88(11), 1322-1335. doi: 10.2522/ptj.20080008

Calapai, G. (2008). European legislation on herbal medicines: a look into the future. *Drug Safety*, 31, 428-431.

Calixto, J.B. (2000). Efficacy, safety, quality control, marketing and regulatory guidelines for herbal medicines (phytotherapeutic agents). *Brazilian Journal of Medical and Biological Research*, 33, 179-189.

Campbell-Tofte, J. I., Mølgaard, P., Josefsen, K., Abdallah, Z., Hansen, S. H., Cornett, C., & Winther, K. (2011). Randomized and double-blinded pilot clinical study of the safety and anti-diabetic efficacy of the *Rauwolfia-Citrus* tea, as used in Nigerian traditional medicine. *Journal of Ethnopharmacology*, 133(2), 402-411.

Catelani, G., D'Andrea, F., Griselli, A., Guazzelli, L., Legnani, L., & Toma, L. (2008). A new stereoselective approach to a selectively protected derivative of d-pinitol and its evaluation as α -l-rhamnopyranose mimetic. *Tetrahedron Letters*, 49(29), 4534-4536.

Centers for Disease Control and Prevention. (2011), National diabetes fact sheet: national estimates and general information on diabetes and prediabetes in the United States, 2011. Atlanta, GA: U.S. Department of Health and Human Services, Centers for Disease Control and Prevention.

Chadwick, W.A., Roux, S., van de Venter, M., Louw, J. & Oelofsen, W. (2007). Anti-diabetic effects of *Sutherlandia frutescens* in Wistar rats fed a diabetogenic diet. *Journal of Ethnopharmacology*, 109, 121-127.

Chemistry 108B, Chapter 26-Lecture 2. (2005). [<http://www.chemistry.ucsc.edu/teaching/Spring05/Chem108B/Chapter26-Lecture2.pdf>]. accessed: 15.06.2016

- Daneman, D. (2006). Type 1 diabetes. *The Lancet*, 367(9513), 847-858.
- Davies, A., Christensen, M., Horowitz, J. F., Plein, S., Hellerstein, M. K., Ostlund Jr, R. E. (2000). Effect of pinitol treatment on insulin action in subjects with insulin resistance. *Diabetes Care*, 23, 1000-1005.
- Deal, G. (2010). Overview and update on EU directive 2004/24/EC: traditional herbal medicines. *Reg. Rapporteur*, 7(7/8): 10-14.
- Department of Health. (2006). Guidelines for Good Practice in the Conduct of Clinical Trials with Human Participants in South Africa. Department of Health: Pretoria, South Africa. 0955406464
- Depaula, A. L., Macedo, A. L., Rassi, N., Vencio, S., Machado, C. A., Mota, B. R., Silva, L. Q., Halpern A., & Schraibman V. (2008). Laparoscopic treatment of metabolic syndrome in patients with type 2 diabetes mellitus. *Surgical Endoscopy*, 22(12), 2670-2678.
- Deshpande, A.D., Harris-Hayes, M. & Schootman, M. (2008). Epidemiology of diabetes and diabetes-related complications. *Physical Therapy*, 88, 1254-1264.
- Einhorn, D., Rendell, M., Rosenzweig, J., Egan, J. W., Mathisen, A. L., Schneider, R. L., & Pioglitazone 027 Study Group. (2000). Pioglitazone hydrochloride in combination with metformin in the treatment of type 2 diabetes mellitus: a randomized, placebo-controlled study. *Clinical therapeutics*, 22(12), 1395-1409.
- Eldin, I. M. T., Ahmed, E. M., & HM, A. E. (2009). Clinical hypoglycemic effects of *Allium cepa* (red onion) in type 2 diabetic patients. *Gezira Journal of Health Sciences*, 5(2).
- Emanuel, E. J., & Miller, F. G. (2001). The ethics of placebo-controlled trials—a middle ground. *New England Journal of Medicine*, 345(12), 915-919.
- Emanuel, E. J., Wendler, D., & Grady, C. (2000). What makes clinical research ethical? *JAMA*, 283(20), 2701-2711.
- Emanuel, E. J., Wendler, D., & Grady, C. (2000). What makes clinical research ethical?. *JAMA*, 283(20), 2701-2711.
- Ernst, E. (2003). Serious adverse effects of unconventional therapies for children and adolescents: a systematic review of recent evidence. *European Journal of Pediatrics*, 162, 72-80. doi:10.1007/s00431-002-1113-7
- European Medicines Agency, (2006a). Guideline on quality of herbal medicinal products/traditional herbal medicinal products. *EMEA/CVMP/814/00 Rev 1*.
- European Medicines Agency, (2006b). Guidelines on specifications: test procedures and acceptance criteria for herbal substances, herbal preparations and herbal medicinal products/traditional herbal medicinal products. *EMEA/CVMP/815/00 Rev1*.
- Evans, J. S. B. (2009). How many dual-process theories do we need? One, two, or many? DOI:10.1093/acprof:oso/9780199230167.003.0002

- Evans, W. C. (2002) Trease and Evans Pharmacognosy. Edinburgh: Saunders.
- Fakhoury, W. K., LeReun, C., & Wright, D. (2010). A meta-analysis of placebo-controlled clinical trials assessing the efficacy and safety of incretin-based medications in patients with type 2 diabetes. *Pharmacology*, 86(1), 44-57.
- Faleschini, M. T., Myer, M. S., Harding, N., & Fouche, G. (2013). Chemical profiling with cytokine stimulating investigations of *S. frutescens* LR (Br.) (Fabaceae). *South African Journal of Botany*, 85, 48-55.
- Feld, S. (2002). The American Association of Clinical Endocrinologists medical guidelines for the management of diabetes mellitus: the AACE system of intensive diabetes self-management-2002 update. *Endocrine Practice*, 8(Supplement 1), 40-82.
- Fernandes, A. C., Cromarty, A. D., Albrecht, C., & van Rensburg, C. E. J. (2004). The antioxidant potential of *Sutherlandia frutescens*. *Journal of Ethnopharmacology*, 95(1), 15.
- Firenzuoli, F. (2006). Important bias in the Astragalus meta-analysis. *Journal of Clinical Oncology*, 24, 3215-3216. doi:10.1200/JCO.2006.06.2695
- Firenzuoli, F., & Gori, L. (2007). Herbal Medicine Today: Clinical and Research Issues. *Evidence-based Complementary and Alternative Medicine*, 4, 37-40. doi:10.1093/ecam/nem096
- Food and Drug Administration (FDA). (2010). Overview of dietary supplements. Retrieved from <http://www.fda.gov/food/dietarysupplements/consumerinformation>.
- Gagnier, J.J., Boon, H., Rochon, P., Moher, D., Barnes, J., Bombardier, C., (2006). Reporting Randomized, Controlled Trials of Herbal Interventions: An Elaborated CONSORT Statement. *Ann. Intern. Med.* 144, 364–367. doi:10.7326/0003-4819-144-5-200603070-00013
- García-Pérez, L. E., Álvarez, M., Dilla, T., Gil-Guillén, V., & Orozco-Beltrán, D. (2013). Adherence to therapies in patients with type 2 diabetes. *Diabetes Therapy*, 4, 175-194. doi:10.1007/s13300-013-0034-y
- Gericke, N., Albrecht, C. F., Van Wyk, B., Mayeng, B., Mutwa, C., & Hutchings, A. (2001). *Sutherlandia frutescens*. *Australian Journal of Medical Herbalism*, 13(1), 9-15.
- Ghorbani, A. (2013). Best herbs for managing diabetes: a review of clinical studies. *Brazilian Journal of Pharmaceutical Sciences*, 49, 413-422.
- Ghorbani, A., & Rakhshandeh, H. (2012). The most effective herbs for diabetes. Mashhad, Iran: Mashhad University of Medical Sciences Publisher.
- Giordano, J., Engebretson, J., & Garcia, M. K. (2005). Challenges to complementary and alternative medical research: Focal issues influencing integration into a cancer care model. *Integrative cancer therapies*, 4(3), 210-218.

Gkaliagkousi, E., Ashish, S., & Alberto, F. (2007). Pharmacological and non-pharmacological treatment of endothelial dysfunction: relevance to diabetes. *The British Journal of Diabetes and Vascular Disease*, 7, 5-10.

Hosseini, S., Jamshidi, L., Mehrzadi, S., Mohammad, K., Najmizadeh, A. R., Alimoradi, H., & Huseini, H. F. (2014). Effects of *Juglans regia L.* leaf extract on hyperglycemia and lipid profiles in type two diabetic patients: A randomized double-blind, placebo-controlled clinical trial. *Journal of Ethnopharmacology*, 152(3), 451-456. DOI: 10.1016/j.jep.2014.01.012

Huseini, H. F., Larijani, B., Heshmat, R. A., Fakhrzadeh, H., Radjabipour, B., Toliat, T., & Raza, M. (2006). The efficacy of *Silybum marianum (L.) Gaertn. (Silymarin)* in the treatment of type II diabetes: a randomized, double-blind, placebo-controlled, clinical trial. *Phytotherapy Research*, 20(12), 1036-1039.

Hussain, S. A. R. (2007). *Silymarin* as an adjunct to glibenclamide therapy improves long-term and postprandial glycemic control and body mass index in type 2 diabetes. *Journal of medicinal food*, 10(3), 543-547.

International Diabetes Federation. (2012). IDF Diabetes Atlas [homepage on the Internet]. [Cited 2012 Mar 18]. Available from: <http://www.idf.org/diabetesatlas>.

International Diabetes Federation. (2015). IDF Diabetes Atlas 7th ed., Brussels, Belgium; International Diabetes Federation. Retrieved from <<http://www.diabetesatlas.org>>.

Jayawardena, M. H. S., De Alwis, N. M. W., Hettigoda, V., & Fernando, D. J. S. (2005). A double blind randomized placebo controlled cross-over study of a herbal preparation containing *Salacia reticulata* in the treatment of type 2 diabetes. *Journal of Ethnopharmacology*, 97(2), 215-218.

Jocham, G., & Ullsperger, M. (2008) Neuropharmacology of performance monitoring. *Neuroscience and Behavioral Reviews*, 33, 48-60.

Johnson, Q., Syce, J., Nell, H., Rudeen, K., & Folk, W. R. (2007). A randomized, double-blind, placebo-controlled trial of *Lessertia frutescens* in healthy adults. *PloS clinical trials*, 2(4), e16

Kajimoto, O., Kawamori, S., Shimoda, H., Kawahara, Y., Hirata, H., & Takahashi, T. (2000). Effects of a diet containing *Salacia reticulata* on mild type 2 diabetes in humans. A placebo-controlled, cross-over trial. *Nippon Eiyo Shokuryo Gakkaishi= Journal of the Japanese Society of Nutrition and Food Science*, 53(5), 199-205.

Karlberg, J. P. E., & Speers, M. A. (Eds.). (2010). Reviewing clinical trials: a guide for the ethics committee (pp. 23-27). Hong Kong: Karlberg.

Karlon H. J., Mohsen B., & Cherpitel J. C. (2001). Alcohol, Tobacco, and Drug Use and the Onset of Type 2 Diabetes Among Inner-city Minority Patients. *The Journal of the American Board of Family Practice*, 14(6), 430-436.

- Kim, M. J., Yoo, K. H., Kim, J. H., Seo, Y. T., Ha, B. W., Kho, J. H., & Chung, C. H. (2007). Effect of pinitol on glucose metabolism and adipocytokines in uncontrolled type 2 diabetes. *Diabetes Research and Clinical Practice*, 77(3), S247-S251.
- Lavalle-González, F. J., Januszewicz, A., Davidson, J., Tong, C., Qiu, R., Canovatchel, W., & Meininger, G. (2013). Efficacy and safety of canagliflozin compared with placebo and sitagliptin in patients with type 2 diabetes on background metformin monotherapy: a randomised trial. *Diabetologia*, 56(12), 2582-2592.
- Lawrence, W., Conrad, S., & Moore, A. S. (2012). Type 2 diabetes: Growing to epic proportions. *Nursing management*, 43(1), 20-25.
- Li, J. W.-H., & Vederas, J.C. (2009). Drug Discovery and Natural Products: End of an Era or an Endless Frontier? *Science*, 325, 161-165. doi:10.1126/science.1168243
- Loomans, C.J., de Koning, E.J., Staal, F.J., Rookmaaker, M.B., Verseyden, C., de Boer, H.C., Verhaar, M.C., Braam, B., Rabelink, T.J., & van Zonneveld, A.-J. (2004). Endothelial progenitor cell dysfunction a novel concept in the pathogenesis of vascular complications of type 1 diabetes. *Diabetes*, 53, 195-199.
- Loya, A. M., González-Stuart, A., & Rivera, J. O. (2009). Prevalence of polypharmacy, polyherbacy, nutritional supplement use and potential product interactions among older adults living on the United States-Mexico border. *Drugs & Aging*, 26(5), 423-436.
- Lu, F. R., Shen, L., Qin, Y., Gao, L., Li, H., & Dai, Y. (2008). Clinical observation on *trigonella foenum-graecum* L. total saponins in combination with sulfonylureas in the treatment of type 2 diabetes mellitus. *Chinese Journal of Integrative Medicine*, 14, 56-60.
- MacKenzie, J., Koekemoer, T., van de Venter, M., Dealtry, G., & Roux, S. (2009). *Sutherlandia frutescens* limits the development of insulin resistance by decreasing plasma free fatty acid levels. *Phytotherapy Research*, 23(11), 1609-1614.
- Mahady, G. B. (2001). Global harmonization of herbal health claims. *Journal of Nutrition*, 131, 1120S-1123S.
- Mang, B., Wolters, M., Schmitt, B., Kelb, K., Lichtinghagen, R., Stichtenoth, D. O., & Hahn, A. (2006). Effects of a *cinnamon* extract on plasma glucose, HbA1c, and serum lipids in diabetes mellitus type 2. *European Journal of Clinical Investigation*, 36(5), 340-344.
- Marić, A. (2010). Metformin—more than ‘gold standard’ in the treatment of type 2 diabetes mellitus. Department of Internal Medicine, Čakovec County Hospital, Čakovec, Croatia.
- Mathew, P. T., & Augusti, K. T. (1974). Hypoglycaemic effects of onion, *Allium cepa* Linn. on diabetes mellitus—a preliminary report. *Indian Journal of Physiology and Pharmacology*, 19(4), 213-217.
- Miller, F. G., Emanuel, E. J., Rosenstein, D. L., & Straus, S. E. (2004). Ethical issues concerning research in complementary and alternative medicine. *JAMA*, 291(5), 599-604.

- Miller, F.G. 2004. Ethical Issues Concerning Research in Complementary and Alternative Medicine. *JAMA*, 291, 599. doi:10.1001/jama.291.5.599
- Mills, E., Cooper, C., Seely, D., & Kanfer, I. (2005). African herbal medicines in the treatment of HIV: Hypoxis and *Sutherlandia*. An overview of evidence and pharmacology. *Nutrition Journal*, 4(1),1.
- Modak, M., Dixit, P., Londhe, J., Ghaskadbi, S., Devasagayam, T.P.A. (2007). Indian herbs and herbal drugs used for the treatment of diabetes. *Journal of Clinical Biochemistry and Nutrition*, 40, 163-173.
- Möller, H. J. (2011). Effectiveness studies: advantages and disadvantages. *Dialogues in clinical neuroscience*, 13(2), 199.
- Moshe, D. (1998). A biosystematic study of the genus *Sutherlandia* R.Br. (*Fabaceae*, *Galegeae*). MSc Thesis (Botany), University of Johannesburg.
- Moshe, D., van der Bank, H., van der Bank, M., & van Wyk, B. E. (1998). Lack of genetic differentiation between 19 populations from seven taxa of *Sutherlandia* Tribe: *Galegeae*, *Fabaceae*. *Biochemical Systematics and Ecology*, 26(6), 595-609.
- Myint, Z., Htun, H. L., Thwin, T., Mar, T. T., Nwe, M. M., Oo, A. M., Maw, L. Z., Oo, T. K. K., & Kyaw, M. T. (2009). Acute effect of onion (*Allium cepa*) on blood glucose level of diabetic patients. *Myanmar Health Sciences Research Journal*, 21(1), 22-25.
- Nathan, D. M., Buse, J. B., Davidson, M. B., Ferrannini, E., Holman, R. R., Sherwin, R., & Zinman, B. (2009). Medical management of hyperglycaemia in type 2 diabetes mellitus: a consensus algorithm for the initiation and adjustment of therapy. *Diabetologia*, 52(1), 1730.
- Newman, D. J., & Cragg, G. M. (2007). Natural products as sources of new drugs over the last 25 years. *Journal of Natural Products*, 70(3), 461-477.
- Ngcobo, M., Nkala, B., Moodley, I., & Gqaleni, N. (2012). Recommendations for the Development of Regulatory Guidelines for Registration of Traditional Medicines in South Africa. *African Journal of Traditional, Complementary and Alternative Medicines*, 9(1), 59-66.
- NIH. (2014). Insulin Resistance and Prediabetes. NIH Publication No. 14-4893. http://diabetes.niddk.nih.gov/dm/pubs/insulinresistance/Insulin_Resistance_Prediabetes-508.pdf. 29th October 2014
- Ojewole, J.A.O., 2004. Analgesic, anti-inflammatory and hypoglycemic effects of *Sutherlandia frutescens* R.Br. (variety incana E. Mey.) [*Fabaceae*] shoot aqueous extract. *Methods and Findings in Experimental and Clinical Pharmacology* 26, 409-416.
- Ostlund, R. E., & Sherman, W. R. (1996). U.S. Patent No. 5,550,166. Washington, DC: U.S. Patent and Trademark Office.

- Ozougwu, O. (2013). The pathogenesis and pathophysiology of type 1 and type 2 diabetes mellitus. *Journal of Physiology and Pathophysiology*, 4, 46-57. doi:10.5897/JPAP2013.0001
- Pan, S. Y., Zhou, S. F., Gao, S. H., Yu, Z. L., Zhang, S. F., Tang, M. K., ... & Ko, K. M. (2013). New perspectives on how to discover drugs from herbal medicines: CAM's outstanding contribution to modern therapeutics. *Evidence-Based Complementary and Alternative Medicine*, 2013.
- Pradesh, A. (2011). Quality and regulatory affairs of herbal drugs: A world-wide Review. *American Journal of Pharm Research*, 1(5), 389-396.
- Prevoo, D., Swart, P., & Swart, A. C. (2008). The influence of *Sutherlandia frutescens* on adrenal steroidogenic cytochrome P450 enzymes. *Journal of Ethnopharmacology*, 118(1), 118-126.
- Radha, R., & Amrithaveni, M. (2009). Role of medicinal plant *Salacia Reticulata* in the management of type II diabetic subjects. *Ancient Science of Life*, 29(1), 14.
- Ramezani, M. A. J. I. D., Azarabadi, M., Abdi, H., Baher, G., & Huseini, M. (2008). The effects of *Silybum marianum* (L.) Gaertn. seed extract on glycemic control in type II diabetic patient's candidate for insulin therapy visiting endocrinology clinic in baqiyatallah hospital in the years of 2006. *Journal of Medicinal Plants*, 2(26), 79-84.
- Reid, K. A., Maes, J., Maes, A., van Staden, J., De Kimpe, N., Mulholland, D. A., & Verschaeve, L. (2006). Evaluation of the mutagenic and antimutagenic effects of South African plants. *Journal of Ethnopharmacology*, 106(1), 44-50.
- Ritov, V. B., Menshikova, E. V., Azuma, K., Wood, R., Toledo, F. G., Goodpaster, B. H., ... & Kelley, D. E. (2010). Deficiency of electron transport chain in human skeletal muscle mitochondria in type 2 diabetes mellitus and obesity. *American Journal of Physiology-Endocrinology and Metabolism*, 298(1), E49-E58.
- Rodbard, H. W., Blonde, L., Braithwaite, S. S., Brett, E. M., Cobin, R. H., Handelsman, Y., & Mechanick, J. I. (2007). AACE Diabetes Mellitus Clinical Practice Guidelines Task Force American Association of Clinical Endocrinologists medical guidelines for clinical practice for the management of diabetes mellitus. *Endocrine Practice*, 13(Suppl 1), 1-68.
- Rojas, L. B. A., & Gomes, M. B. (2013). Metformin: an old but still the best treatment for type 2 diabetes. *Diabetology & Metabolic Syndrome*, 5(1), 1.
- Rothman, K. J., & Michels, K. B. (1994). The continuing unethical use of placebo controls. *New England Journal of Medicine*, 331(6), 394-398.
- Routledge, P.A. (2008). The European Herbal Medicines Directive: could it have saved the lives of Romeo and Juliet? *Drug Safety*, 31, 416-418.
- Scheen, A.J. (2003). Pathophysiology of Type 2 Diabetes. *Acta Clinica Belgica*, 58, 335-341. doi:10.1179/acb.2003.58.6.001

Schmidt, B., Ribnicky, D. M., Poulev, A., Logendra, S., Cefalu, W. T., & Raskin, I. (2008). A natural history of botanical therapeutics. *Metabolism* 57, S3-S9. doi:10.1016/j.metabol.2008.03.001

Seier, J. V., Mdhluli, M., Dhansay, M. A., Loza, J., Laubscher, R., (2002). A toxicity study of Sutherlandia leaf powder (*Sutherlandia microphylla*) consumption. Final report: April 2002. Medical Research Council of South Africa and National Research Foundation. www.sa-healthinfo.org/traditionalmeds/firststudy.htm

Sharma, A. K., Kumar, R., Mishra, A., & Gupta, R. (2010). Problems associated with clinical trials of *Ayurvedic* medicines. *Revista Brasileira de Farmacognosia*, 20(2), 276-281.

Shaw, J. E., Sicree, R. A. & Zimmet, P. Z. (2010). Global estimates of the prevalence of diabetes for 2010 and 2030. *Diabetes Research and Clinical Practice*, 87(1), 4-14.

Sia, C. (2004). Spotlight on ethnomedicine: usability of *Sutherlandia frutescens* in the treatment of diabetes. *The Review of Diabetic Studies*, 1(3), 145-149.

Sia, C. (2004). Spotlight on ethnomedicine: usability of *Sutherlandia frutescens* in the treatment of diabetes. *The Review of Diabetic Studies*, 1(3), 145.

Skyler, J. S., Skyler, D. L., Seigler, D. E., & O'Sullivan, M. J. (1981). Algorithms for adjustment of insulin dosage by patients who monitor blood glucose. *Diabetes Care*, 4(2), 311-318.

Soltani, R., Gorji, A., Asgary, S., Sarrafzadegan, N., & Siavash, M. (2015). Evaluation of the Effects of *Cornus mas L.* Fruit Extract on Glycemic Control and Insulin Level in Type 2 Diabetic Adult Patients: A Randomized Double-Blind Placebo-Controlled Clinical Trial. *Evidence-Based Complementary and Alternative Medicine*.

Suksomboon, N., Poolsup, N., Boonkaew, S., & Suthisisang, C.C. (2011). Meta-analysis of the effect of herbal supplement on glycemic control in type 2 diabetes *Journal of Ethnopharmacology*, 137, 1328-1333.

Tai, J., Cheung, S., Chan, E., & Hasman, D. (2004). In vitro culture studies of *Sutherlandia frutescens* on human tumor cell lines. *Journal of Ethnopharmacology*, 93(1), 9-19.

Tilburt, J. (2008). Herbal medicine research and global health: an ethical analysis. *Bulletin of the World Health Organization*, 86, 594-599. doi:10.2471/BLT.07.042820

Tuomilehto, J., Lindstorm, J., Eriksson, J. G., Valle, T. T., Hämäläinen, H., Ilanne-Parikka, P., Keinänen-Kiukaanniemi, S., Laakso, M., Louheranta, A., Rastas, M., Salminen, V., & Uusitupa, M.; for the Finnish Diabetes Prevention Study Group. (2001). Prevention of type 2 diabetes mellitus by changes in life-style among subjects with impaired glucose tolerance *New England Journal of Medicine*, 344, 1343-1350.

United States Department of Health and Human Services, Food and Drug Administration. (2006). Guidance for industry on Complementary and Alternative Medicine (CAM) products and their regulation by the Food and Drug Administration: draft guidance.

Van Wyk, B. E., & Albrecht, C. (2008). A review of the taxonomy, ethnobotany, chemistry and pharmacology of *Sutherlandia frutescens* (Fabaceae). *Journal of Ethnopharmacology*, 119(3), 620-629.

Van Wyk, B. E., Oudtshoorn, B. V., & Gericke, N. (1997). Medicinal Plants of South Africa. Briza Publications.

Van Wyk, B., & Gericke, N. (2000). People's plants: A guide to useful plants of Southern Africa. Briza Publications.

Van Wyk, B-E., Wink, M., 2004. Medicinal plants of the world. Briza Publications, Pretoria. ISBN 1 875093 44 3.

Vaz, J. A., & Patnaik, A. (2012). Diabetes mellitus: Exploring the challenges in the drug development process. *Perspectives in Clinical Research*, 3, 109. doi:10.4103/2229-3485.100660

Vlietinck, A., Pieters, L., & Apers, S. (2009). Legal Requirements for the Quality of Herbal Substances and Herbal Preparations for the Manufacturing of Herbal Medicinal Products in the European Union. *Planta Medica*, 75, 683-688. doi:10.1055/s-0029-1185307

Wachtel-Galor, S., & Benzie, I. F. F. (2011). Herbal Medicine: An Introduction to Its History, Usage, Regulation, Current Trends, and Research Needs. In I. F. F. Benzie & S. Wachtel-Galor, (Eds.), *Herbal medicine: Biomolecular and clinical aspects*. Boca Raton, FL: CRC Press/Taylor & Francis.

Whiting, D. R., Guariguata, L., Weil, C., & Shaw, J. (2011). IDF diabetes atlas: global estimates of the prevalence of diabetes for 2011 and 2030. *Diabetes research and clinical practice*, 94(3), 311-321.

Williams, S., Roux, S., Koekemoer, T., van de Venter, M., & Dealtry, G. (2013). *Sutherlandia frutescens* prevents changes in diabetes-related gene expression in a fructose-induced insulin resistant cell model. *Journal of Ethnopharmacology*, 146(2), 482-489.

Wilson, D., Goggin, K., Williams, K., Gerkovich, M. M., Gqaleni, N., Syce, J., Bartman, P., Johnson, Q., & Folk, W. R. (2015). Consumption of *Sutherlandia frutescens* by HIV-Seropositive South African Adults: An adaptive double-blind randomized placebo-controlled trial. *PloS One*, 10(7), e0128522.

Wilson, D., Syce, J. A., & Goggin, K. (2010). A Randomized, Double-blind, Placebo-controlled Study of the Safety and Efficacy of *Lessertia frutescens* (L.) Goldblatt and JC Manning (syn. *S. frutescens* (L.) R. Br.) in HIV-infected South Africans: Protocol.

World Health Organization (1985). Report of a WHO Study Group. Geneva Chapter 2, Diabetes Mellitus Technical Report Series 727.

World Health Organization Regional Office for Africa, (2001). Promoting the role of traditional medicine in health systems. A strategy for the African region. AFR/RC50/9.

World Health Organization Regional Office for Africa, (2004). Guidelines for registration of TM in the WHO African region. AFR/TRM/04.1.

World Health Organization. (1999). Definition, Diagnosis and Classification of Diabetes Mellitus and its Complications. Report of a WHO Consultation. Geneva

World Health Organization. (2005). National Policy on Traditional Medicine and Regulation of Herbal Medicines: Report of a WHO global survey. Geneva, Switzerland: World Health Organization.

World Health Organization. (2011). Diabetes: Key Facts. Geneva, Switzerland: World Health Organization.

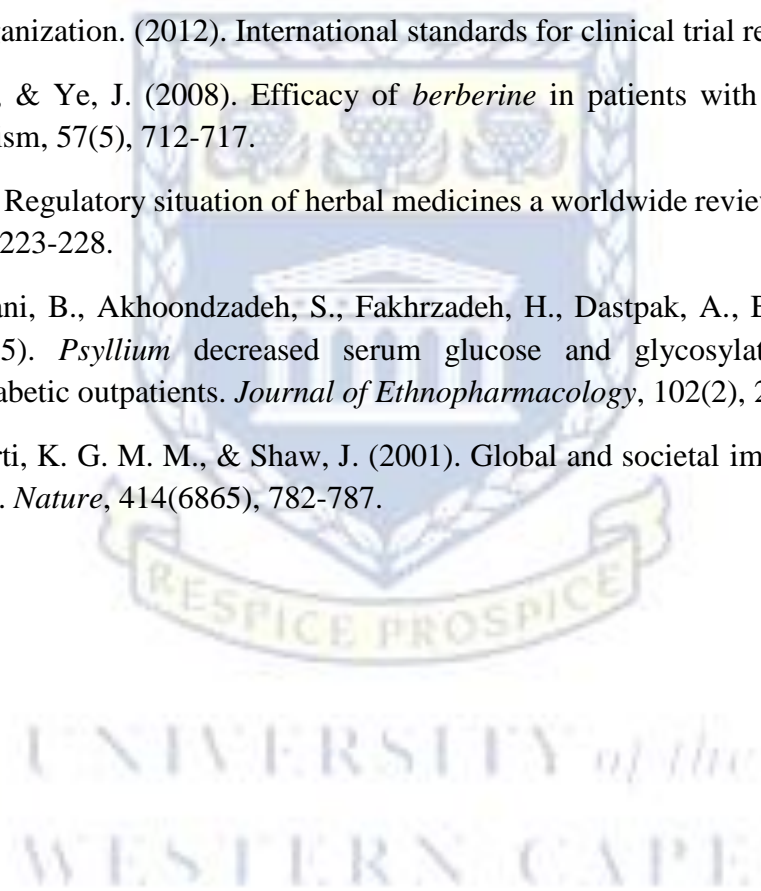
World Health Organization. (2012). International standards for clinical trial registries.

Yin, J., Xing, H., & Ye, J. (2008). Efficacy of *berberine* in patients with type 2 diabetes mellitus. *Metabolism*, 57(5), 712-717.

Zhang, X. (1998). Regulatory situation of herbal medicines a worldwide review. *World Health Organization*, 26, 223-228.

Ziai, S. A., Larijani, B., Akhoondzadeh, S., Fakhrzadeh, H., Dastpak, A., Bandarian, F., & Emami, T. (2005). *Psyllium* decreased serum glucose and glycosylated hemoglobin significantly in diabetic outpatients. *Journal of Ethnopharmacology*, 102(2), 202-207.

Zimmet, P., Alberti, K. G. M. M., & Shaw, J. (2001). Global and societal implications of the diabetes epidemic. *Nature*, 414(6865), 782-787.



APPENDIX 1

Complete Protocol

Protocol Title:	A randomized double-blind placebo-controlled study to investigate the efficacy of <i>Sutherlandia frutescens</i> (<i>subspecies microphylla</i>) in adult type-2 diabetics.
Protocol Number:	UWC 16-002 version: 04
Protocol Date:	17 September 2016
Study Phase:	II
Project Leader (SA):	Prof. J Syce
Protocol Authors:	Mr. Ramadan Swead, Prof J A Syce
Product Quality Investigator:	Prof. J Syce, MPharm, PhD
Sponsor:	TBA
Implemented By:	School of Pharmacy University of Western Cape (UWC), Private Bag X17, Bellville, 7535, South Africa

Confidentiality Agreement

This document is a confidential communication of UWC School of pharmacy. Acceptance of this document constitutes the agreement by the recipient that no unpublished information contained within will be published or disclosed without prior written approval, except that this document may be disclosed to the appropriate Ethics Committee and Regulatory Authority under the condition that they are requested to keep it confidential.

TABLE OF CONTENTS

PAGE

STUDY CONTACT LIST	
LIST OF ABBREVIATIONS AND DEFINITIONS	4
PROTOCOL SYNOPSIS	5
1 INTRODUCTION	9
1.1 Background	9
1.2 Study Rationale	11
1.3 Study Objectives	12
1.4 Study Population	12
1.5 Number of Participant Planned	12
1.6 Inclusion Criteria	12
1.7 Exclusion Criteria	13
1.8 Justification for inclusion and exclusion criteria	13
1.9 Criteria for Discontinuation	14
1.10 Study Endpoints	15
1.11 Safety Endpoints	15
1.12 Primary Safety	15
1.13 Secondary Safety Endpoints	15
1.14 Efficacy Endpoints	15
1.15 Investigational Plan	16
1.16 Study Design	16
1.17 Scheduled Clinic Visits	16
1.18 Visits and Assessments	17
1.19 Measurements at each visit	17
1.20 Specific Detail on Measurements	21
1.21 Adverse Events (AE)	21
1.22 Safety Reporting	23
1.23 Investigational product	23
1.24 Investigational Products and Treatments	23
1.25 Treatment Schedule	23
1.26 Randomization	23
1.27 Study Product Dosing	24
1.28 Study Duration	24
1.29 Identity of Study Products	24
1.30 Storage and Accountability	25
1.31 Allowed Medication	25
1.32 Compliance	25
1.33 Statistics	26
1.34 Statistical Dosing/model	26
1.35 Planned Analyses	26
1.36 Ethics	27
1.37 Ethics Review	27
1.38 Ethical Conduct of the Study	27
1.39 Participants Informed Consent	27
1.40 Data Quality Assurance	28
1.41 Study Time Table and Termination	28
1.42 References	29
1.43 Appendix	31

LIST OF ABBREVIATIONS AND DEFINITIONS

The following abbreviations and specialist terms are used in this study protocol:

Abbreviation	Explanation
<i>S.f</i>	<i>Sutherlandia frutescens</i>
T2DM	Type-2 Diabetes Mellitus
AE	Adverse Event
HbA1c	Glycosylated Hemoglobin (A1C)
FBG	Fasting Blood Glucose
BMI	Body Mass Index
B.i.d	Bis in die = twice daily
CRF	Case Report Form
ECG	Electrocardiogram
Hb	Haemoglobin
Hct	Haematocrit
HDL-C	High Density Lipoprotein –cholesterol
IEC	Independent Ethics Committee
IRB	Independent Review Board
LDL-C	Low Density Lipoprotein -cholesterol
ml	Milliliter
n	Sample Size
MRC	Medical Research Council
RBC	Red Blood Cells
SAE	Serious Adverse Event
SG	Specific Gravity
TTC	Tijger Trial Centre
UWC	University of the Western Cape
WBC	White Blood Cells
GCP	Good Clinical Practice
MCC	Medicinal Control Council

Definitions

Herbal Medicines: Mainly whole, fragmented or cut plants, part of plants in an unprocessed state, usually in dried form, but sometimes fresh. Herbal drugs are precisely defined by the botanical scientific name according to the binomial system (genus, species, variety).

African traditional medicine: The total body of knowledge and techniques for the preparation and use of substances, measures, and practices that are based on the socio-cultural and religious bedrock of African communities. This is founded on personal experience and observations handed down from generation to generation, either verbally or in writing, and are used for the diagnosis, prevention or elimination of imbalances in physical, mental or social well-being.

PROTOCOL SYNOPSIS

TITLE	A randomized double blind placebo controlled study to investigate the efficacy of <i>Sutherlandia frutescens</i> (subspecies <i>microphylla</i>) in adult type-2 diabetes mellitus.
Investigational site:	Clinical Research Centre, the University of Cape Town, L51 Old Main Building, Groote Schuur Hospital, Observatory Road.
Investigators:	TBA
Project Leaders:	Prof. James Syce, Mr. R Swead
Sponsor:	TBA (School of Pharmacy, University of Western Cape)
Clinical Phase:	II
Study number:	UWC 16-002
OBJECTIVES:	<ul style="list-style-type: none"> • To determine the efficacy of <i>Sutherlandia frutescens</i> (subsp. <i>Microphylla</i>) capsules in adult type-2 diabetes mellitus by assessing, HbA1C. • To evaluate the proportion of participants achieving a therapeutic response (HbA1c < 7%) with <i>S. frutescens</i> 1200mg versus placebo after 12 weeks of treatment • To determine the effect of <i>S. frutescens</i> on glycemc control (HbA1c, fasting plasma glucose, etc)
STUDY DESIGN:	6-month randomized double-blind placebo-controlled study. Participants will be screened at Visit 1 and eligible subjects will, at visit 2, be randomized to either <i>Sutherlandia frutescens</i> (subsp. <i>Microphylla</i>) or placebo. 1200mg capsule, b.i.d or a placebo similar appearance.



UNIVERSITY of the
WESTERN CAPE

Participants:	<p>Inclusion criteria</p> <ol style="list-style-type: none"> 1. T2DM men and women between 18–70 years of age on a stable medication. HBA1C >6.5% <8.5% 2. Fasting plasma glucose (FPG) 7.0–14.0 mmol/l 3. Have body weights within 25% of the appropriate weight range 4. Had taken standardized diet control + exercise therapy + stable administration of metformin for more than 3 months before being enrolled. 5. Has a normal ECG as determined by a unit physician. 6. Has not taken any traditional medication for 28 days prior to screening. 7. Be informed of the nature of the study and will give written informed consent. <p>Exclusion criteria</p> <ol style="list-style-type: none"> 1. Type 1 diabetes mellitus, gestational diabetes mellitus and other special types of diabetes mellitus. 2. In the past 3 months, had taken medication to control body weight (including weight-loss drug) or oral administration of any anti-diabetic drugs or insulin except for metformin. 3. Had diabetic ketosis, ketoacidosis. 4. Fasting triglycerides >450 mg/dL (>5.1 mmol/L) 5. Had uncontrolled hypertension (blood pressure be and more than 160/100 mmHg). 6. Intake of anti-diabetic medication within 14 days before the start of the study. 7. Had mental illness, alcohol addiction and /or taken administration of psychoactive drug substance. 8. Smokers, who smoke more than 10 cigarettes per day and cannot, refrain from smoking during the study period. 9. Pregnancy, lactation or being prepared, pregnant woman 10. Had allergic constitution. 11. Unwillingness or inability to follow the procedures outlined in the protocol. 12. Any disease or condition which might compromise the haematopoietic, renal, endocrine, pulmonary, central nervous, cardiovascular systems.
PRODUCT TO BE EVALUATED	<p>Test: <i>Sutherlandia frutescens</i> 1,200mg /day, capsules.</p> <p>Reference: Placebo capsules</p>
DURATION OF STUDY	6 months ±30 days
ENDPOINTS:	<p>Primary End Point: primary endpoints are defined as weight loss and change in HbA1c levels comparing between the treatment groups.</p> <ol style="list-style-type: none"> 3. Vital parameters: pulse rate, blood pressure, respiratory rate, weight, and height will be recorded at the baseline visit only. 4. Adverse events and serious adverse events. To follow a non- exhaustive list and any adverse event that patients reported will be tracked. These events are as follows: <ul style="list-style-type: none"> h) CNS (e.g. headaches, nervousness, dizziness) i) GIT (e.g. diarrhea, gastritis, constipation, stomach cramps, nausea). j) Infections k) Allergy (e.g. dermatitis). l) Appetite (e.g. increase, decrease). m) General (e.g. iliac pain). n) Others <p>The following primary efficacy precautions will be measured per the flowchart on the page.</p> <ol style="list-style-type: none"> 4. Full count blood and differential count.

	<p>5. Physical examination.</p> <p>6. 12-lead electrocardiogram.</p> <p>Secondary Endpoints: The following endpoints will be measured at the baseline, week 4, week 12, and week 24;</p> <p>Efficacy Endpoints: The following efficacy endpoints will be obtained at the designated study visits:</p> <ol style="list-style-type: none"> 5. Medical history. 6. Medical symptoms. 7. Alcohol/substance use.
SAMPLE SIZE:	100 subjects
STATISTICAL ANALYSIS:	<p>An intention to the treatment approach will be used. The primary endpoint for efficacy studies is the end result of treatment with either the placebo or the drug. The data will be analysed as a within patient comparison of treatments using the t-test for paired differences. The t-test will be also used to test for the period effect and the carry over effect.</p>



UNIVERSITY of the
WESTERN CAPE

FLOWCHART

Study stage	Treatment period											
	-1 screening	0 baseline	2	4	6	8	12	16	20	24	28 follow up if applicable	
Informed consent	x											
General Information	x											
Vital Signs	x	x	x	x	x	x	x	x	x	x	x	x
Inclusion/exclusion criteria	x	x										
Review of medical symptoms	x	x	x	x	x	x	x	x	x	x		
Blood and urine routine												
Liver and kidney function	x	x										
Twelve-lead ECG	x					x	x			x		
Fasting blood glucose	x	x	x	x	x	x	x	x	x	x	x	x
Investigational drug dose adjustment	x	x										
Fasting insulin	x	x		x		x	x		x	x	x	x
HbA1c	x	x										
Blood lipid	x	x										
Physical examination	x					x	x			x	x	
Height	x											
Pregnancy test	x	x		x	x	x	x	x	x	x		
Allocation of subject number	x	x										
Collection of urine sample	x	x		x		x	x			x		
Collection of blood sample	x	x		x		x	x			x		
Background/moderating variables												
Evaluation of traditional medicine diabetes symptoms	x	x										
hypoglycemia	x	x			x	x				x		
Adverse events	x	x	x	x	x	x	x	x	x	x	x	x
Appetite			x									
Release of investigational drug	x	x	x			x			x	x		
Recovery of investigational drug	x	x	x	x		x	x	x	x	x		
Alcohol / substance use		x		x	x		x	x	x	x		
Study medication Dispensed/returned (D/R)		D	D/ R	D/ R	D/ R	D R	D/ R	D/ R	D/ R	D/ R	R	
Drug adherence – pill counts			x	x	x	x	x	x	x	x		
Unannounced phone pill counts				x	x							

1. Introduction

1.1 Background

Diabetes is a serious global health issue. According to the World Health Organisation (WHO), the prevalence of diabetes worldwide in 2030 is estimated to 366 million. Type-2 diabetes mellitus account for approximately 90 to 95% of all cases (Rodbard *et al.*, 2007), and that 80% of these diabetics live in low- and middle-income countries (Olokoba, *et al.*, 2012 & IDF, 2012). Approximately 6.6% of the world's population aged between 20 and 79 years has diabetes type-2, and this estimation is projected to increase to 7.8% by 2030 (Atlas, D. 2006 & Fakhoury *et al.*, 2010). Type-2 diabetes is associated with serious long-term microvascular and macrovascular complications such as cardiovascular disease, neuropathy, and diseases of the eyes, which in turn, increase morbidity, mortality, and health care costs (Cade, 2008). There is no cure for diabetes, but the condition can, however, be controlled with a lifelong commitment to blood sugar monitoring, weight management, nutrition and exercise making it very restrictive. General treatment of type-2 diabetes includes; healthy eating, regular exercise, diabetes medication or insulin therapy and blood sugar monitoring. Metformin is used in patients with type-2 diabetes and is considered as the “gold standard” (Maric, 2010). According to (Scarpello, J, H., 2008), metformin is now established as a first-line antidiabetic therapy for the management of type-2 diabetes. According to Ghorbani (2013), several herbs have been recommended for treating diabetes however, the recommendations are based on animal studies and limited pieces of evidence exist about their clinical usefulness.

Sutherlandia frutescens (subspecies microphylla) (L.) R.Br. is thought to be among the most profound and useful medicinal plants in Southern Africa (van Wyk & Gericke, 2000). Tinctures, infusions, and decoctions of the leaves and young stems have been widely used in the Cape Region since the value of this herbal medicine was first discovered by the Khoi, San and Nama people (van Wyk, van Oudtshoorn & Gericke, 2000). Furthermore, the North Sotho name, *Lerumo-lamadi*, means the spear for the blood which refers to the value of *S. frutescens* as a powerful blood-purifier that acts as an all-purpose tonic. Over centuries, this medicinal plant has been indigenously used to treat a variety of conditions including type-2 diabetes mellitus (van Wyk & Gericke, 2000). The medicinal value of the plant has been ascribed to a number of the significant constituents of this phytotherapy, including pinitol, γ -amino butyric acid (GABA), asparagine, sugaryl (SU1) and also L-canavanine. Pinitol, which is a known anti-diabetic agent (Davis *et al.*, 2000 & Larner *et al.*, 1998), has been isolated from *S. frutescens* leaves. Pinitol is claimed to have a clinical application in treating due to diabetes patients (van

Wyk & Gericke, 2000). (Singh, *et al.*, 2001). *Sutherlandia frutescens* extracts have been shown to possess antioxidant properties in a cell-free system that may partially explain the plant's anti-inflammatory effects, and other recent studies have described below.

A recently completed study on vervet monkeys (*Chlorocebus Aethiops*) conducted by the South Africa Medical Research Council (MRC), reported that up to nine times the recommended dose of *S.frutescens* (81mg/kg body weight per day for three months) resulted in no significant changes to relevant haematological, biochemical and physiological parameters. It was concluded that *S. frutescens* are non-toxic in vervet monkeys (www.sahealthinfo.org/traditionalmeds/firststudy.htm, 2002). Results of Phase I double-blind placebo-controlled study conducted by The International Center for Indigenous Phytotherapy Studies (TICIPS) through the South African Herbal Science and Medicine Institute (SAHSMI) at the University of the Western Cape to evaluate the safety of *S. frutescens* in 25 healthy adult volunteers have just been made available (Johnson, Q, Syce, *et al.*, 2007). This randomized double blind placebo controlled study randomized 12 healthy subjects to the treatment arm where they received 400mg *S. frutescens* leaf powder capsules twice a day and 13 subjects to the control arm where they received an identical placebo capsule twice per day. Participants were seen at monthly visits throughout the 3-month study period. The subjects provided blood and urine samples for haematology/biochemistry (i.e., RBC, WBC, serum analysis, serum proteins) and urinalysis (i.e., SG, pH, protein, glucose, ketone, bilirubin, blood, & urobilinogen), completed a 12-lead ECG, and reported any adverse events at each visit. Results indicate that the 800mg per day dose was well tolerated by study participants and resulted in no significant changes to relevant haematological, biochemical or physiological parameters (Johnson, Q, Syce, *et al.*, 2007).

The National Center for Complementary and Alternative Medicine (NCCAM), National Institutes of Health (NIH) and Department of Medicine at Edendale Hospital, Pietermaritzburg, South Africa to evaluate the efficacy of *S. frutescens* in HIV patients. In Stage 1, 56 participants were randomized to *S. frutescens* 400, 800 or 1,200 mg twice daily or matching placebo for 24 weeks (Wilson, Douglas, *et al.* 2015). In Stage 2, 77 additional participants were randomized to either 1,200 mg *S. frutescens* or placebo. In the final analysis data from Stage 1 and Stage, 2 were combined such that 107 participants were analysed (54 in the *S. frutescens* 1,200 mg arm and 53 in the placebo arm). *S. frutescens* did not change HIV viral load, and CD4 T-lymphocyte count was similar in the two arms at 24 weeks; however, mean and total burden of infection

(BOI; defined as days of infection-related events in each participant) was greater in the *S. frutescens* arm: mean (SD) 5.0 (5.5) vs. 9.0 (12.7) days ($p = 0.045$), attributed to two tuberculosis cases in subjects taking isoniazid preventive therapy (IPT), Wilson, Douglas, *et al.* (2015).

1.2 Study Rationale

Type-2 diabetes is the most common form of diabetes, accounting for 90 - 95% of cases (Centers for Disease Control, 2011). It has been further estimated that diabetes prevalence for South Africans is at 6.46% for adults aged between 20-79 years. *S. frutescens* is thought to be among the most efficacious plants used in Southern African traditional medicine. The plant has been used in indigenous settings to treat diabetes, as well as inflammatory conditions. It's possible that *S. frutescens* may have useful effects in adult type-2 diabetes.

There has been few properly conducted clinical trials performed on traditionally used herbal medicines in South Africa and *S. frutescens* is no exception. Nevertheless, this herbal medicine is being used widely without any apparent recorded adverse effects and has the potential to be of immense benefit.

Given the widespread indigenous use of *S. frutescens* as a phytotherapeutic and its non-toxic effects are shown in the pre-clinical studies in a primate, it is logical to assess its efficacy, during chronic use, in clinical trials.

In both the traditional and commercial settings, the normal recommended dose of *L. frutescens* is approximately 600 to 800 mg daily. However, it is known that in the traditional setting doses are substantially higher than this frequently used and/or the *L. frutescens* combined with other plant medicines. The outcomes of the completed phase I study showed that 800mg (12 mg/kg for an average 67 kg human) daily doses of *L. frutescens* leave powder produced no serious effect while doses of 3x and 9x (i.e. 81 mg/kg body weight) the equivalent human dose also produced no adverse effects in the vervet monkey. To optimize the chance of clearly establishing whether *L. frutescens* is safe and efficacious or not in the traditional setting, a dose-escalating study to determine the bioavailability for the safe and effective use of the product would thus be well justified.

2. Study Objective

2.1 Primary objective

- To determine the efficacy of *S. frutescens* in type-2 diabetes in adults.

Hypothesis: The number and category of adverse events in participants taking *S. frutescens* will not differ significantly from the subjects taking the placebo.

2.2 Secondary objective

- To determine the effect Hb1Ac that *L. frutescens* has on type-2 diabetes in adults versus the placebo.
- To evaluate the proportion of participants achieving a therapeutic response (HbA1c < 7%) with *S. f* 1,200mg versus placebo after 12 weeks of treatment
- To evaluate the effect of *Sutherlandia frutescens* on glycemic control (HbA1c, fasting plasma glucose, etc)

3. Study Population

3.1 Number of Participants Planned

100 subjects are to be enrolled in the study. The following criteria have to be fulfilled at both the Screening (Visit 0) and Randomisation (Visit 1):

3.2 Inclusion Criteria

A subject will be eligible for inclusion in this study only if all of the following criteria are met:

1. T2DM man and woman between 18-70 years of age on a stable medication.
2. HBA1C >6.5% <8.5%
3. Fasting plasma glucose (FPG) 7.0-14.0 mmol/l
4. Have body weights within 25% of the appropriate weight range
5. Had taken standardized diet control + exercise therapy + stable administration of metformin for more than 3 months before being enrolled.
6. Has a normal ECG as determined by a unit physician.
7. Has not taken any traditional medication for 28 days prior to screening.
8. Be informed of the nature of the study and will give written informed consent.

3.3 Exclusion Criteria

A subject will not be eligible for inclusion in this study if any of the following criteria apply:

1. Type1 diabetes mellitus, gestational diabetes mellitus and other special types of diabetes mellitus.
2. In the past 3 months, had taken medication to control body weight (including weight-loss drug) or oral administration of any anti-diabetic drugs or insulin except for metformin.
3. Had diabetic ketosis, ketoacidosis.
4. Fasting triglycerides >450mg/dL (>5.1 mmol/L)
5. Had uncontrolled hypertension (blood pressure be and more than 160/100 mmHG).
6. Intake of anti-diabetic medication within 14 days before the start of the study.
7. Had mental illness, alcohol addiction and /or taken administration of psychoactive drug substance.
8. Smokers, who smoke more than 10 cigarettes per day and cannot, refrain from smoking during the study period.
9. Pregnancy, lactation or being prepared pregnant woman
10. Had allergic constitution.
11. Unwillingness or inability to follow the procedures outlined in the protocol
12. Any disease or condition which might compromise the haematopoietic, renal, endocrine, pulmonary, central nervous, cardiovascular systems.

3.4 Justification for inclusion and exclusion criteria

The criterion is set to minimize the risk to the participants, to ensure a subject population that will enable the investigation of the set objectives. In order to provide equal opportunity for inclusion and not to exclude subjects that may be usual users of the herbal medicine. Because this is the first formal efficacy study on *Sutherlandia frutescens* in type-2 diabetes mellitus, children are excluded from it; their inclusion may, however, be considered in subsequent studies.

3.5 Criteria for Discontinuation

Participation in the study is voluntary. Study participants are free to withdraw from the study at any time. If they decide to withdraw from the study, they should inform the study doctor immediately. The doctor should not be upset or penalize the study in any way. Withdrawal from the study should not impact their future care in any way. If a study participant withdraws

from the study, the study data collected before the withdrawal may still be processed along with other data collected as part of the study.

In addition, the following circumstances may lead to a participant's discontinuation from the study:

- Medical reasons, including pregnancy.
- Progression of disease.
- Incorrect enrollment or randomization of the subject.
- Noncompliance of a study subject; e.g. a study participant not taking their medication or not regularly keeping to the study appointment, or if they don't follow the doctor's instructions.
- If the sponsor stops the study.
- If the Medicines Control Council or the Research Ethics Committee stops or suspends the investigation.

For subjects withdrawn from the study, the measurements and assessment should be performed as done at each visit. Adverse events should be followed up and the study medication should be returned by the subject.

4. Study Endpoints

4.1 Safety Endpoints

4.1.1 Primary Safety

The following primary safety endpoints will be measured at each study visit from the beginning of screening to week 24.

Adverse events will be measured at all study visits following the start of the study medication.

1. Vital parameters; pulse rate, blood pressure, respiratory rate, oral temperature, weight and height will be recorded at the baseline visit only.
2. Adverse events and serious adverse events with start and stop dates. Given the finding of the earlier phase I clinical study, the following list represented potential AEs that might be observed. However, this is not an exhaustive list and any adverse event that patients reported will be tracked.
 - a) CNS (e.g. headaches, nervousness, dizziness).
 - b) GIT (e.g. diarrhea, gastritis, constipation, stomach cramps, nausea).
 - c) Infections.
 - d) Allergy (e.g. dermatitis).
 - e) Appetite (e.g. increase, decrease).

- f) General (e.g. iliac pain).
- g) Others.

The following primary safety will be measured per the flowchart on page

1. Full blood count and differential count.
2. Physical examination.
3. 12-lead electrocardiogram.

4.1.2 Secondary Safety Endpoints

The following secondary endpoint will be measured at baseline, week 4, week12 and week24.

4.2 Efficacy Endpoints

The following efficacy endpoint will be measured at week 24.

5. Investigational Plan

5.1 Study Design

This study will utilise a participant and assessor blinded-placebo randomised controlled trial design to evaluate the effectiveness of *Sutherlandia frutescens* in the treatment of adult type-2 diabetics. A parallel design with allocation on a 1:1 ratio will be used. Eligible participants will be randomised to either a 6 months *Sutherlandia frutescens* intervention or placebo. A pharmaceutical company will be consulted to develop the drug in collaboration with researchers at the University of the Western Cape. Study staff, the principle investigator, and outcome assessors will not have access to the randomization (treatment) code. Access will only be in cases were a serious adverse event occurs. In such cases only in exceptional circumstances, the code will be broken when knowledge of the allocation is vital for treating the patient.

5.2 Scheduled Clinic Visits

Visit number	Type of visit	Day number(days between visits)
Visit 0 screening	Information , screening, and consent	Day -7
Visit 1 Baseline	Randomization & 1 st Drug issue visit	Day 0 (7± 2 days after visit 0)
Visit 2 Week 2	After 2 weeks treatment	Day 14 (14 ± 2 days after visit 1)
Visit 3 Week 4	After 4 weeks treatment	Day 28 (28 ± 2 days after visit 1)
Visit 4 Week 8	After 8 weeks treatment	Day 56 (56 ± 3 days after visit 1)
Visit 5 Week 12	After 12 weeks treatment	Day 84 (84 ± 3 days after visit)
Visit 6 Week 16	After 16 weeks treatment	Day 112 (112 ± 5days after visit)
Visit 7 Week 20	After 20 weeks treatment	Day 140 (140 ± 5 days after visit)
Visit 8 Week 24	After 24 weeks treatment	Day 168 (168 ± 5days after visit)

5.3 Visits and Assessments

Study participants will be assessed for adverse events potentially attributable to *Sutherlandia frutescens* at screening, baseline, week 2, week 4, week 8, week 12, week 16, week 20, week 24, and 4 weeks after the last dose, if applicable.

5.4 Measurements at each visit

5.4.1 At visit 0: Screening Visit

The participants will be examined within 14 days prior to the first ingestion of the investigational products to assess their eligibility to participate. Each participant will consent in writing to the study before the start of the investigational procedures. The informed consent form will also include the study information leaflet.

The investigations and examinations will include:

- Medical history, including the history of past use of medications, demographics (date of birth, sex, race), contraception history and alcohol and tobacco consumption patterns.
- Vital signs: pulse, systolic and diastolic blood pressure, respiratory rate, body weight and oral temperature. Blood pressure will be taken in the same arm consistently.

- Physical examination including assessment of general appearance, cardiovascular, neurological, lungs, mouth, throat and abdomen and measurement of height. The examination will be made in accordance with the normal clinical routines at the clinical study center.
- Review of medical signs and symptoms.
- Urine β -HCG pregnancy test in non-hysterectomized volunteers. Fertile females must have a negative pregnancy test.
- ECG: standard 12-lead.
- Review of concomitant medications.
- Laboratory tests: Haematology, serum biochemistry, and urine analysis.
- Investigator confirms that study participant fulfills inclusion criteria for the study.

5.4.2 Baseline at visit 1: Day 1 - Randomization & 1st Drug Issue.

- Study participants will be seen at the investigational center and investigator will confirm that study subjects fulfill inclusion criteria for the study.
- Vital signs: pulse, systolic and diastolic blood pressure, respiratory rate, body weight and oral temperature. Blood pressure will be taken in the same arm consistently.
- The pre-dose safety assessments and eligibility checks will be recorded on the relevant pages in the individual CRFs.
- Review of medical signs and symptoms.
- Urine β -HCG pregnancy test.
- Concomitant medication assessment.
- Assign study Participant Identification Number (PIN).
- The randomized subject will be issued with 1 month's supply of study medication, instructed how to take the medication and given the next appointment date. Subjects should start the first dose at the recommended time (ca: 06:00 – 10:00) on the day of drug issue or start the next day. In addition, they will receive a 1-week emergency supply of study medication to be used if needed before the next study visit. This will be checked at each study visit and re-supplied as needed.
- Storage of serum and urine for analysis for *Sutherlandia* metabolites.

5.4.3 At visit 2: Treatment and Assessment (Week 2)

For this visit the examinations and investigations will include:

- A collection of returned study medication, pill count and reissue of months study medication.
- Vital signs: systolic and diastolic blood pressure, pulse, respiratory rate, weight and oral temperature. Blood pressure will be consistently taken in the same arm.
- Study medication compliance will be checked and discussed (esp. with respect to incidence and type of reported adverse events).
- Adverse event and concomitant medication assessment.
- Review of medical signs and symptoms.

5.4.4 At visit 3: Treatment and assessment (Week 4)

For this visit the investigations and examinations will include:

- A collection of returned study medication and pill count.
- Study medication compliance will be checked and discussed (esp. with respect to incidence and type of reported adverse events).
- Vital signs: systolic and diastolic blood pressure, pulse, respiratory rate, weight and oral temperature. Blood pressure will be consistently taken in the same arm.
- Urine β -HCG pregnancy test.
- Laboratory tests: Haematology, serum biochemistry, and urine analysis.
- Review of medical signs and symptoms.
- The subject will be issued with 1 month's supply of study medication; instructions on how to take the medication will be reviewed and given next appointment date.
- Adverse events and concomitant medication assessment.
- Between Visit 3 and 4, adherence to study medication will be assessed by unannounced phone contact and pill count.

5.4.5 Visit 4: Treatment and assessment (Week 8)

For this visit the examinations and investigations will include:

- A collection of returned study medication and pill count.
- Vital signs: systolic and diastolic blood pressure, pulse, respiratory rate, weight and oral temperature. Blood pressure will be consistently taken in the same arm.

- Study medication compliance will be checked and discussed (esp. with respect to incidence and type of reported adverse events).
- Review of medical signs and symptoms.
- Laboratory tests: Haematology, serum biochemistry, and urine analysis
- Adverse event and concomitant medication assessment.
- Urine β -HCG pregnancy test.
- The subject will be issued with 1 month's supply of study medication; instructions on how to take the medication will be reviewed and given next appointment date.
- Between Visit 4 and 5, adherence to study medication will be assessed by unannounced phone contact and pill count.

5.4.6 Visit 5: Treatment and assessment (Week 12)

For this visit the examinations and investigations will include:

- A collection of returned study medication and pill count.
- Vital signs: systolic and diastolic blood pressure, pulse, respiratory rate, weight and oral temperature. Blood pressure will be consistently taken in the same arm.
- Study medication compliance will be checked and discussed (esp. with respect to incidence and type of reported adverse events).
- Review of medical signs and symptoms.
- Laboratory tests: Haematology, serum biochemistry, and urine analysis
- Adverse event and concomitant medication assessment.
- Urine β -HCG pregnancy test.
- The subject will be issued with 1 month's supply of study medication; instructions on how to take the medication will be reviewed and given next appointment date.
- Physical examination.
- ECG: standard 12-lead.

5.4.7 Visits 6 & 7: Treatment and assessment (Week 16 and Week 20)

For this visit the examinations and investigations will include:

- A collection of returned study medication and pill count.
- Study medication compliance will be checked and discussed (esp. with respect to incidence and type of reported adverse events).

- Vital signs: systolic and diastolic blood pressure, pulse, respiratory rate, weight and oral temperature. Blood pressure will be consistently taken in the same arm.
- Urine β -HCG pregnancy test.
- Review of medical signs and symptoms.
- Adverse event and concomitant medication assessment.
- The subject will be issued with 1 month's supply of study medication; instructions on how to take the medication will be reviewed and given next appointment date.

5.4.8 Visits 8: Treatment and assessment (Week 24)

For this visit the examinations and investigations will include:

- A collection of returned study medication and pill count.
- Study medication compliance will be checked and discussed (esp. with respect to incidence and type of reported adverse events).
- Vital signs: systolic and diastolic blood pressure in sitting (after 5 minutes rest), pulse, respiratory rate, weight and oral temperature. Blood pressure will be consistently taken in the same arm.
- Review of medical signs and symptoms.
- Laboratory tests: Haematology, serum biochemistry, and urine analysis.
- Urine β -HCG pregnancy test.
- Physical examination.
- ECG: standard 12-lead.
- Assess Quality of Life and potential mediators and background/moderating variables.
- Adverse event and concomitant medication assessment.
- A collection of returned dosettes.
- Storage of serum and urine for analysis for *Sutherlandia* metabolites.

Note: For any subject in which any of the above tests indicated levels outside the normal range or an (s)AE is ongoing, a follow-up visit (visit 9) will be scheduled for an appropriate time (1 month after completion of study participation or withdrawal). Specific tests will be repeated at this visit, as needed.

5.5 Specific Details on Measurements

5.5.1 Adverse Events (AE)

An adverse event is the development of an undesirable medical condition - e.g. symptoms or abnormal results of an investigation - or the deterioration of a pre-existing medical condition

(not relevant in this study). AE's will be collected by means of a standard question: "Have you had any health problems since the previous visit?" AE's will be recorded at every visit. Spontaneously reported AE's and/or observed AE's and the subject's response to this question will be recorded on the AE form with information about seriousness, the action was taken, date of onset and recovery, maximum intensity, and outcome. The subjects will be asked to assess the intensity of the reported Adverse Event according to the following scale:

- Mild = awareness of sign or symptom, but easily tolerated
Moderate = discomfort sufficient to cause interference with normal activities
Severe = incapacitating, with the inability to perform normal activities.

A Serious Adverse Event is an adverse event occurring during any phase of the study and at any dose of the investigational product or placebo, which fulfils one or more of the following criteria:

- Results in death;
- Is immediately life-threatening;
- Subject hospitalization;
- Results in persistent or significant disability or incapacity.

The causality of Serious Adverse Events (i.e. the relationship to study treatment) will be assessed by the investigators, who in completing the relevant Case Report Form must answer 'yes' or 'no' to the question "Do you consider that there is a reasonable possibility that the event may have been caused by the study medication?" The following factors should be considered when deciding if there is a "reasonable possibility" that an Adverse Event may have been caused by the investigational product.

- Time course of events and exposure to suspect drug – did the AE occur in a reasonable temporal relationship to the administration of suspect drug?
- De-challenge experience – did the AEs resolve or improve on stopping or reducing the dose of the suspect drug?
- Re-challenge experience - did the AEs reoccur if the suspected drug was reintroduced after having stopped?
- Laboratory tests – has a specific laboratory investigation confirmed the relationship?

- No alternative cause, the AEs cannot be reasonably explained by aetiology such as an underlying disease (not previously present), other drugs or environmental factors.

There would not be a “reasonable possibility” of causality if none of the above criteria apply or where there is evidence of exposure and a reasonable time course, but any de-challenge is negative or there is another more likely cause of the AEs.

In this study, the Adverse Events will be noted from the interview at the time of visit and from the daily input in the diary.

5.5.2 Safety Reporting

All serious adverse events will be reported to the Sponsor, the South African Medicines Control Council, National Center for Complementary and Alternative Medicine, and the Research Ethics Committee within 24 hours of the study team becoming aware of the event. All adverse events will be reported in summary form during the study to the Sponsor and the South African Medicines Control Council. The Data and Safety Monitoring Board affiliated with UWC will be asked to undertake an independent review of the adverse events occurring in the study if three or more serious adverse events occur during the course of the study. Unblinding will occur after written notification by the Principal Investigator or the Data and Safety Monitoring Board.

6. Investigational product

6.1 Investigational Products and Treatments

All study treatment supplied are manufactured, tested and released according to current Good Manufacturing Practice Guidelines (GMP).

6.2 Treatment Schedule

6.2.1 Randomization

Participant identification numbers (PIN) will be randomly linked to either *S. frutescens* dose (1200mg/day) or placebo by the study statistician. At the baseline visit, eligible participants will be sequentially assigned PIN and to study arm. All clinical staff will be blinded to the study drug / placebo assignment, and the study statistician will be located off-site at the University of the Western Cape. Participants will be unblinded only after written request by the Lead Clinical Investigator or as required by the Date and Safety Monitoring Board.

6.2.2 Study Product Dosing

Participants will take either the test product or placebo in a capsule daily.

6.2.3 Study Duration

Participants will take either the test product or placebo for 24 weeks.

6.3 Identity of Study Products

The study product is a standard (20mm x 6mm) capsule filled with *Sutherlandia* leaf powder. Before use, the consistency of the product will be validated by pharmaceutically evaluating samples of the capsules for mass and content uniformity and dissolution profile (using pinitol as markers). The product will then be repacked (in quantities of 60) into containers identical to that to be used for the placebo capsules. Identically looking placebo product will be prepared by filling the capsules with a mixture comprising appropriate amounts of lactose, starch and a small amount of dried spinach leaf powder. The placebo products and test product validation will be done in the Pharmaceutics Laboratory, School of Pharmacy, and UWC. Assaying for the pinitol will be done using a validated LCMS assay based on the method of Petritis *et al* (2000) and a gas chromatographic method based on the method of Lein, *et al.* (2002), respectively. A certificate of analysis will be available before the test product is used.

Study code: UWC 16-002	Visit no: 8
Investigator: N/A	Subject no: 100
Investigational product: <i>Sutherlandia frutescens</i> or placebo	
Dosage form: 1capsule twice daily	
Store below 30°C	Exp Date: N/A
For clinical study use only	

6.4 Storage And Accountability

All study drugs/or medications must be kept in a secure place under adequate storage conditions – protected from moisture and light. Records of dispensing and returns will be maintained by the investigational site. The participant/or subject must return all unused study medication for each treatment period to the investigational site for reconciliation.

6.5 Allowed Medication

Preferably no other medicine should be taken by the participants. The use of any incidental medication (e.g. mild analgesics, oral contraceptives, etc) will be recorded at visit 1.

6.6 Compliance

Compliance will be assessed by counting remaining pills returned by participants at each follow-up visit and by two unannounced pill counts by phone during the 6-month trial.

Study code: UWC 16-002	Investigator: N/A	
Subject Number: 100	Week Number: N/A	Bottle Number: N/A
Capsules containing <i>Sutherlandia</i> or Placebo		
This Investigational Product Should be stored out of the Reach of Children and Used by Study Participant Only.		
Directions: Put one capsule from each bottle into each a.m. box and one capsule from each bottle into each p.m. box.		
Store below 30°C	For clinical study use only	Batch number: N/A

7. Statistics

7.1 Statistical Dosing/model

Appropriate descriptive analyses will be performed to examine distributional characteristics for collected measures, and to summarize changes over time as a function of group assignment.

7.2 Planned Analyses

A single statistical analysis (of one treatment group vs placebo) will be performed at the end of the study. An intention to treat (ITT) approach will be followed, i.e. statistical analysis of safety will be based on data from all patients who were randomized and from whom meaningful data were collected. Data will be displayed graphically for visual inspection. Descriptive statistics will be presented as means, SEM, and ninety percent confidence levels of the means. Additional analyses will be conducted to explore possible effects of mediators and moderators on outcomes.

Baseline characteristics: The subject disposition will be summarized. The demographic, background and baseline data will be presented descriptively.

Analysis of safety: Adverse Events, as reported throughout the course of the study will be listed individually, per treatment group. Pre-study and post-study findings of physical examination, vital sign variables, laboratory variables (haematology, clinical chemistry, and urinalysis), and 12-lead ECG will be listed individually and summarized; values outside the normal range will be listed. Protocol violations will be listed per subject, describing the nature of the violation. Subjects failing to complete the study (as well as the times and reasons for discontinuation) will be displayed.

Analysis of constituent levels: The active constituents obtained during the treatment will be compared to that found before the start of study treatment, and the averaged levels obtained in the trial medication and placebo groups will be compared and analyzed using an analysis of variance model.

8. Ethics

8.1 Ethics Review

The study to be initiated, the protocol and informed consent and participant information form should be reviewed and received approval by an Independent Ethics Committee (IEC) and Regulatory Authority. The agreement of the final study protocol, including the final version of the Subject Information and Consent Forms, must be in writing form by the IEC and MCC before enrolment of any participant.

The Clinical Trial Manager (Principal Investigator) is responsible for informing the IEC of any serious adverse events (SAEs) and amendment to the protocol as per regulatory requirement.

8.2 Ethical Conduct Of The Study

The study will be performed in accordance with the ethical principles of the Declaration of Helsinki and will be consistent with Good Clinical Practice and applicable regulatory requirements.

8.3 Participants Informed Consent

First, to any participation in the clinical study, subjects should provide written informed consent. The Investigator will ensure that the subject is given full and adequate oral and written information about the nature, purpose, possible risk and benefit of the study. Subjects must also be notified that they are free to discontinue from the study at any time. The subject should be given the opportunity to ask questions and allowed time to consider the information provided.

The subjects signed and dated informed consent must be obtained before conducting any study specific procedure. The investigator must store the original, signed Written Informed Consent Form and a copy must be given to the subject.

9. Data Quality Assurance

Monitoring and auditing procedure, as determined will be followed, in order to comply with Good Clinical Practice and to ensure acceptability of the study data for intentional publication purposes.

Data from the study will be collected in CRFs. Data editing will be performed at the trial center, comparing source and CRF entries. Data will be entered in a blind mode.

During the study, an independent monitor will visit the investigational site to confirm that the facilities remain acceptable, that the investigational team is adhering to the protocol and that data are being accurately recorded in the CRFs. Source data verification (a comparison of the data in the CRF with the subject's laboratory test results and other source documents) will also be performed.

Authorized representatives of the regulatory authority (e.g. MCC) may visit the center (site) to perform inspections, including source data verification.

Clean File for the final database will be declared once all the data has been entered and a quality check on a sample of the data has been performed. The database will be locked after Clean File has been declared and data will be extracted for statistical analysis. Treatment codes will not be broken until the clean file has been performed.

Study committee meetings will be held as needed prior to or during the study. The medical, nursing and other staff involved in the study will receive proper education/information on how to conduct the study according to the protocol.

10. Study Time Table and Termination

Application:

First, subject in:

Last patient out:

Clean File:

Study Report:

11. References to protocol

1. Atlas, D. (2006). International diabetes federation. *Press Release, Cape Town, South Africa, 4*.
2. Cade, W.T. (2008). Diabetes-Related Microvascular and Macrovascular Diseases in the Physical Therapy Setting. *Phys Ther*, 88(11): 1322–1335. doi: 10.2522/ptj.20080008.
3. Centers for Disease Control and Prevention. (2011). National diabetes fact sheet: national estimates and general information on diabetes and prediabetes in the United States, 2011. *Atlanta, GA: US Department of Health and Human Services, Centers for Disease Control and Prevention, 201(1)*.
4. Davis A., Christiansen M, Horowitz JF, Klein S, Hellerstein MK, and Ostlund RE Jr. (2000). *Diabetes Care*, 23(7), 1000 -1005.
5. Fakhoury, W. K., LeReun, C., & Wright, D. (2010). A meta-analysis of placebo-controlled clinical trials assessing the efficacy and safety of incretin-based medications in patients with type 2 diabetes. *Pharmacology*, 86(1), 44-57.
6. Farnsworth N. R., (1995). Alfalfa pills and autoimmune diseases. *Am J Clin Nutr*: 62(5); 1026-1028.
7. Ghogomu, T.A. (2002). Evaluation of *Sutherlandia frutescens* for anti-cancer activity. MS thesis, UWC.
8. Ghorbani, A. (2013). Best herbs for managing diabetes: A review of clinical studies *Brazilian Journal of Pharmaceutical Sciences*, 49, (3).
9. International Diabetes Federation (2012). IDF Diabetes Atlas [homepage on the Internet]. [Cited 2012 Mar 18]. Available from: <http://www.idf.org/diabetesatlas>.
10. Johnson, Q., Syce, J., Nell, H., Rudeen, K., & Folk, W. R. (2007). A randomized, double-blind, placebo-controlled trial of *Lessertia frutescens* in healthy adults. *PloS clinical trials*, 2(4), e16.
11. Larner, J., Allan G., Kessler, C., Reamer, P., Gunn, R. and Huang, L.C. (1998) Phosphoinositol g; ycan-derived mediators and insulin resistance. Prospects for diagnosis and therapy. *Journal of Basic and Clinical Physiology and Pharmacology*: 9(2-4), 127-137.
12. Marić, A. (2010). Metformin—more than ‘GOLD STANDARD’ in the treatment of type 2 diabetes mellitus. *Diabetologia Croatica*, 39(3).
13. Rodbard, Helena, et al. "American Association of Clinical Endocrinologists medical guidelines for clinical practice for the management of diabetes mellitus." *Endocrine*

- practice 13. Supplement 1 (2007): 1-68. Rosenthal, G., In: *Frontiers and New Horizons in Amino Acid Research*, K Takai, (Ed), Elsevier, 1992.
14. Scarpello, J. H., & Howlett, H. C. (2008). Metformin therapy and clinical uses. *Diabetes and Vascular Disease Research*, 5(3), 157-167.
 15. Singh R.K., Pandey B.L., Tripathi, M. and Pandey, V.B. (2001). Anti-inflammatory effect of (+)-pinitol. *Fitoterapia*: 72(2); 168-170.
 16. Van Wyk, B. & Gericke, N. (2000). *People's Plants*, Briza Publications, SA.
 17. Van Wyk, B., Van Oudtshoorn, B. and Gericke, N. (2000). *Medicinal Plants of South Africa*, 2nd Ed, Briza Publications, SA.
 18. Wilson, D., Goggin, K., Williams, K., Gerkovich, M. M., Gqaleni, N., Syce, J., & Folk, W. R. (2015). Consumption of *Sutherlandia frutescens* by HIV-Seropositive South African Adults: An adaptive double-blind randomized placebo-controlled trial. *PloS one*, 10(7), e0128522.
 19. www.sahealthinfo.org/traditionalmeds/firststudy.htm. (2002). A toxicity study of *Sutherlandia leaf powder (Sutherlandia frutescens, sub-species microphylla)* consumption. Medical Research Council, Tygerberg, South Africa.
 20. Olokoba, A. B., Obateru, O. A., & Olokoba, L. B. (2012). Type 2 diabetes mellitus: a review of current trends. *Oman Med J*, 27(4), 269-273.

APPENDICES

DECLARATION OF HELSINKI

Adopted by the 18th WMA General Assembly, Helsinki, Finland, June 1964, and amended by the 29th WMA General Assembly, Tokyo, Japan, October 1975 35th WMA General Assembly, Venice, Italy, October 1983 41st WMA General Assembly, Hong Kong, September 1989 48th WMA General Assembly, Somerset West, Republic of South Africa, October 1996 and the 52nd WMA General Assembly, Edinburgh, Scotland, October 2000

Note of Clarification on Paragraph 29 added by the WMA General Assembly, Washington 2002

INTRODUCTION

1. The World Medical Association has developed the Declaration of Helsinki as a statement of ethical principles to provide guidance to physicians and other participants in medical research involving human subjects. Medical research involving human subjects includes research on identifiable human material or identifiable data.
2. It is the duty of the physician to promote and safeguard the health of the people. The physician's knowledge and conscience are dedicated to the fulfillment of this duty.
3. The Declaration of Geneva of the World Medical Association binds the physician with the words, "The health of my patient will be my first consideration," and the International Code of Medical Ethics declares that "A physician shall act only in the patient's interest when providing medical care which might have the effect of weakening the physical and mental condition of the patient."
4. Medical progress is based on research which ultimately must rest in part on experimentation involving human subjects.
5. In medical research on human subjects, considerations related to the well-being of the human subject should take precedence over the interests of science and society.
6. The primary purpose of medical research involving human subjects is to improve prophylactic, diagnostic and therapeutic procedures and the understanding of the aetiology and pathogenesis of the disease. Even the best-proven prophylactic, diagnostic, and therapeutic methods must continuously be challenged through research for their effectiveness, efficiency, accessibility, and quality.
7. In current medical practice and in medical research, most prophylactic, diagnostic and therapeutic procedures involve risks and burdens.
8. Medical research is subject to ethical standards that promote respect for all human beings and protect their health and rights. Some research populations are vulnerable and need special protection. The particular needs of the economically and medically disadvantaged must be recognized. Special attention is also required for those who cannot give or refuse consent for themselves, for those who may be subject to giving consent under duress, for those who will not benefit personally from the research and for those for whom the research is combined with care.

9. Research Investigators should be aware of the ethical, legal and regulatory requirements for research on human subjects in their own countries as well as applicable international requirements. No national ethical, legal or regulatory requirement should be allowed to reduce or eliminate any of the protections for human subjects set forth in this Declaration.

B. BASIC PRINCIPLES FOR ALL MEDICAL RESEARCH

10. It is the duty of the physician in medical research to protect the life, health, privacy, and dignity of the human subject.
11. Medical research involving human subjects must conform to generally accepted scientific principles, be based on a thorough knowledge of the scientific literature, other relevant sources of information, and on adequate laboratory and, where appropriate, animal experimentation.
12. Appropriate caution must be exercised in the conduct of research which may affect the environment, and the welfare of animals used for research must be respected.
13. The design and performance of each experimental procedure involving human subjects should be clearly formulated in an experimental protocol. This protocol should be submitted for consideration, comment, guidance, and where appropriate, approval to a specially appointed ethical review committee, which must be independent of the investigator, the sponsor or any other kind of undue influence. This independent committee should be in conformity with the laws and regulations of the country in which the research experiment is performed. The committee has the right to monitor ongoing trials. The researcher has the obligation to provide monitoring information to the committee, especially any serious adverse events. The researcher should also submit to the committee, for review, information regarding funding, sponsors, institutional affiliations, other potential conflicts of interest and incentives for subjects.
14. The research protocol should always contain a statement of the ethical considerations involved and should indicate that there is compliance with the principles enunciated in this Declaration.
15. Medical research involving human subjects should be conducted only by scientifically qualified persons and under the supervision of a clinically competent medical person. The responsibility for the human subject must always rest with a medically qualified person and never rest on the subject of the research, even though the subject has given consent.
16. Every medical research project involving human subjects should be preceded by careful assessment of predictable risks and burdens in comparison with foreseeable benefits to the subject or to others. This does not preclude the participation of healthy volunteers in medical research. The design of all studies should be publicly available.
17. Physicians should abstain from engaging in research projects involving human subjects unless they are confident that the risks involved have been adequately assessed and can be satisfactorily managed. Physicians should cease any investigation if the risks are

found to outweigh the potential benefits or if there is conclusive proof of positive and beneficial results.

18. Medical research involving human subjects should only be conducted if the importance of the objective outweighs the inherent risks and burdens to the subject. This is especially important when the human subjects are healthy volunteers.
19. Medical research is only justified if there is a reasonable likelihood that the populations in which the research is carried out stand to benefit from the results of the research.
20. The subjects must be volunteers and informed participants in the research project.
21. The right of research subjects to safeguard their integrity must always be respected. Every precaution should be taken to respect the privacy of the subject, the confidentiality of the patient's information and to minimize the impact of the study on the subject's physical and mental integrity and on the personality of the subject.
22. In any research on human beings, each potential subject must be adequately informed of the aims, methods, sources of funding, any possible conflicts of interest, institutional affiliations of the researcher, the anticipated benefits and potential risks of the study and the discomfort it may entail. The subject should be informed of the right to abstain from participation in the study or to withdraw consent to participate at any time without reprisal. After ensuring that the subject has understood the information, the physician should then obtain the subject's freely-given informed consent, preferably in writing. If the consent cannot be obtained in writing, the non-written consent must be formally documented and witnessed.
23. When obtaining informed consent for the research project the physician should be particularly cautious if the subject is in a dependent relationship with the physician or may consent under duress. In that case, the informed consent should be obtained by a well-informed physician who is not engaged in the investigation and who is completely independent of this relationship.
24. For a research subject who is legally incompetent, physically or mentally incapable of giving consent or is a legally incompetent minor, the investigator must obtain informed consent from the legally authorized representative in accordance with applicable law. These groups should not be included in research unless the research is necessary to promote the health of the population represented and this research cannot instead be performed on legally competent persons.
25. When a subject deemed legally incompetent, such as a minor child, is able to give assent to decisions about participation in research, the investigator must obtain that assent in addition to the consent of the legally authorized representative.
26. Research on individuals from whom it is not possible to obtain consent, including proxy or advance consent, should be done only if the physical/mental condition that prevents obtaining informed consent is a necessary characteristic of the research population. The specific reasons for involving research subjects with a condition that renders them unable to give informed consent should be stated in the experimental protocol for consideration and approval of the review committee. The protocol should state that

consent to remain in the research should be obtained as soon as possible from the individual or a legally authorized surrogate.

27. Both authors and publishers have ethical obligations. In the publication of the results of research, the investigators are obliged to preserve the accuracy of the results. Negative as well as positive results should be published or otherwise publicly available. Sources of funding, institutional affiliations and any possible conflicts of interest should be declared in the publication. Reports of experimentation not in accordance with the principles laid down in this Declaration should not be accepted for publication.

C. ADDITIONAL PRINCIPLES FOR MEDICAL RESEARCH COMBINED WITH MEDICAL CARE

28. The physician may combine medical research with medical care, only to the extent that the research is justified by its potential prophylactic, diagnostic or therapeutic value. When medical research is combined with medical care, additional standards apply to protect the patients who are research subjects.
29. The benefits, risks, burdens and effectiveness of a new method should be tested against those of the best current prophylactic, diagnostic, and therapeutic methods. This does not exclude the use of placebo, or no treatment, in studies where no proven prophylactic, diagnostic or therapeutic method exists. See footnote.
30. At the conclusion of the study, every patient entered into the study should be assured of access to the best proven prophylactic, diagnostic and therapeutic methods identified by the study.
31. The physician should fully inform the patient which aspects of the care are related to the research. The refusal of a patient to participate in a study must never interfere with the patient-physician relationship.
32. In the treatment of a patient, where proven prophylactic, diagnostic and therapeutic methods do not exist or have been ineffective, the physician, with informed consent from the patient, must be free to use unproven or new prophylactic, diagnostic and therapeutic measures, if in the physician's judgement it offers hope of saving life, re-establishing health or alleviating suffering. Where possible, these measures should be made the object of research, designed to evaluate their safety and efficacy. In all cases, new information should be recorded and, where appropriate, published. The other relevant guidelines of this Declaration should be followed.

FOOTNOTE: NOTE OF CLARIFICATION ON PARAGRAPH 29 of the WMA DECLARATION OF HELSINKI

The WMA hereby reaffirms its position that extreme care must be taken in making use of a placebo-controlled trial and that in general this methodology should only be used in the absence of existing proven therapy. However, a placebo-controlled trial may be ethically acceptable, even if proven therapy is available, under the following circumstances:

- Where for compelling and scientifically sound methodological reasons its use is necessary to determine the efficacy or safety of a prophylactic, diagnostic or therapeutic method; or

- Where a prophylactic, diagnostic or therapeutic method is being investigated for a minor condition and the patients who receive placebo will not be subject to any additional risk of serious or irreversible harm.

All other provisions of the Declaration of Helsinki must be adhered to, especially the need for appropriate ethical and scientific review.

The Declaration of Helsinki (Document 17.C) is an official policy document of the World Medical Association, the global representative body for physicians. It was first adopted in 1964 (Helsinki, Finland) and revised in 1975 (Tokyo, Japan), 1983 (Venice, Italy), 1989 (Hong Kong), 1996 (Somerset-West, South Africa) and 2000 (Edinburgh, Scotland). Note of clarification on Paragraph 29 added by the WMA General Assembly, Washington



UNIVERSITY *of the*
WESTERN CAPE

12. APPENDICES

PATIENT INFORMATION LEAFLET AND INFORMED CONSENT

PATIENT INFORMATION LEAFLET AND INFORMED CONSENT

Patient identification number: / Patient initials:

Title of the study: A randomized double-blind placebo-controlled study to investigate the efficacy of *Sutherlandia frutescens* (*subspecies microphylla*) in adult type-2 diabetics.

Protocol number: UWC16-002 version 04

Sponsor: UWC 16-002 version: 04

Name of Principal Investigator: N/A

Name of Project Leader: Prof. James Syce, Mr. R Swead

Address of site/institution where the study will be conducted: Clinical Research Centre, the University of Cape Town, L51 Old Main Building, Groote Schuur Hospital, Observatory Road. And UWC, Student Health Centre, Bellville, Cape Town, South Africa).

INTRODUCTION

You are being invited to take part in a clinical research study sponsored by (x)

Before you decide if you want to join the study you need to know what the study is about. This form tells you about the study and about your rights if you decide to join the study. Please ask the study staff questions at any time.

Before deciding to take part in the study, it is important for you to understand why the research is done and what will happen to you. This information sheet will provide you with information about this study and your rights as a research subject so that you can decide if you want to take part. Please take the time to read this information carefully and ask the investigator when anything is not clear, or if you would like more information.

What is the purpose of this study?

The purpose of this study is to determine the efficacy and safety of the popular plant medicine *Sutherlandia frutescens*. The latter is the study drug, a term that may also be used later on in this document or study. *Sutherlandia frutescens* is also known as Cancer bush in English, Kankerbos in Afrikaans, Unwell in Zulu, etc. We need to accurately establish its efficacy and

safety and to do so in adult's type-2 diabetes mellitus subjects. That is thus the aim of this study.

In this study *Sutherlandia frutescens* (*subsp microphyllin*) in capsule form or a similar looking capsule without the plant (i.e. a placebo) will be given to 100 adult's type-2 diabetes patients. They will take the medicines orally twice daily for 24 weeks while being monitored for any effects of this plant medicine.

The study will be done at Clinical Research Centre, the University of Cape Town, L51 Old Main Building, Groote Schuur Hospital, Observatory Road. & UWC, Student Health Centre, Bellville, Cape Town, South Africa).

Do I have to participate in the study?

You can choose to be in this study or you can choose not to be in this study. Either choice is alright. You have the right to leave this study at any time for any reason. If you choose not to be in this study, you will not lose any of your health care rights. You will still get the care you need. If you decided to participate you need to show that you are willing to join the study by signing at the bottom of this form.

If you withdraw from the study the data collected up to the point of withdrawal will be analyzed for the purpose of the study. If you decide to participate, you will be given this information sheet to keep and be asked to provide your signature indicating your consent.

What will happen to me if I participate?

If you decide to take part you must sign the consent form and will then first be examined to see if you meet all the requirements for participation in the study. These examinations and investigations (at visit 0) will include the taking of a medical history, including history of past use of medications; a physical examination; the withdrawal of 50 ml (approximately 10 teaspoons) of blood for laboratory tests; the recording of an ECG (i.e. an electrocardiogram or recording to check the activity of the heart) and the collection of urine for laboratory test and, if you are female, the collection of urine for a pregnancy test. Samples of blood and urine will be stored in a freezer for testing; including tests to see if *Sutherlandia* is presented.

If you qualify for inclusion you will thereafter, within 14 days of the first examination, be issued (at visit 1) with study medication that you must take as follows: 1 capsule daily in the morning with or after food. There will, however, be 2 study medications, *Sutherlandia frutescens*, and placebo capsules, which will be identical in appearance and, you may receive

one or the other. The allocation of the capsules is done by random selection and neither you nor the study doctor will know on which capsule you are until the study is completed. You have a one in two chance of being given the study drug or placebo. When you start on study (visit1) you will be issued with a month's supply of medication. You will have a check-up every 2 weeks initially for the first two visits (visit 2 and 3) and thereafter every 4 weeks for 24 weeks (visit 4-8) each counted. twice during the study, you will be called on your telephone at home and asked to count the number of capsules you have at home. At the end of the study, you will be asked to bring your unused capsules back to the clinic.

You will also be asked to call or SMS the research staff if you feel unwell. The staff will then call you back every 2 days to find out how you are feeling and what kind of medications you are taking. When you come to the study site for your next visit you will be asked to bring your clinic card with you so the doctor can read the notes made by your clinic nurse or doctor.

You will have to take the study medication for 24 weeks. During this time you will also be asked to come to the research center for 7 visits. At the last visit (8) you may be asked to come for a final visit four weeks after you have taken the last dose of the *Sutherlandia* or placebo. This means that the study will last between 24 and 28 weeks.

During the subsequent visits (**visits 2 to 8**), you will again be examined by the study doctor and asked you if you have experienced any medical problems since the last visit. The same examinations and investigations done during the visit 1 (i.e. the recording of an ECG, withdrawal of 50 ml (810 teaspoons) of blood, etc) will be done. You will not receive any medication at the last visit (**visit 8**) when the study ends.

What do I have to do?

As a subject in this study, you are responsible for:

- keeping all clinic appointments;
- taking the study medication as explained by the study doctor;
- returning the filled-out cards at each visit;
- telling your study doctor before taking any other medication;
- telling your study doctor as soon as possible if there is a change in your health;
- ensuring, if you are a woman of childbearing age, that you take all the needed precautions not to fall pregnant while on this study; and
- returning all used and unused medication and the packaging it came in at each visit

- If you experience any injuries and need to see a doctor, you must also tell that doctor that you are participating in this research study.

What risks or discomforts might occur if I participate?

As with any medication, you may experience some side-effects whilst taking the study drug. The aim of this study is indeed to see if *Sutherlandia frutescens* does indeed cause adverse effects.

So far it has been reported that the study drug (*Sutherlandia*) only produced occasional incidents of mild loose stool, mild constipation, dry mouth and, rarely, dizziness in very frail patients.

Several chemical compounds have however been isolated from *Sutherlandia* and these may produce some effects. Most of these would most likely be beneficial and the reason(s) why this plant may be effective in some diseases. But, some of these potential effects (e.g. lowering of blood glucose, enhancing of autoimmune diseases) may, in some individuals, also be adverse or unwanted. We must be open to the possibility that they can occur. In this study, we will specifically monitor the levels of some of these active substances to see if they may be implicated in any adverse effects of the plant medicine.

Many of the procedures to be used in this study may cause you some discomforts (e.g. the withdrawal of blood may cause slight bruising or possible fainting, there may be discomfort with a recording of the ECG, etc), but these will be no more than that experienced in a typical medical examination.

If you are a woman of childbearing potential, you may take part in the study only if you use a medically accepted method of contraception (birth control). ***Medically accepted methods of birth control include*** the use of oral, intramuscular or transcutaneous contraceptive medication, intrauterine devices, surgical methods i.e. hysterectomy or sterilization and, at the discretion of the clinical investigator a few other methods (e.g. barrier methods, vasectomy in a regular male partner, etc). The study doctor will discuss this with you at the first visit. In addition, you will have a pregnancy test in your blood at the first visit and each time a blood sample is taken. If the pregnancy test is positive, the study doctor will immediately withdraw your medication. You will be asked to sign the supplementary informed consent form to show that you understand your responsibilities should you become pregnant.

What are the possible benefits of participating?

You may or may not receive any direct benefit from taking the study medication, but all the regular visits and examinations to monitor your health will be provided free of charge. Further, the information we get from this study will extend our existing knowledge and will help to decide if this medication is safe to use in other people such as you.

Will there be any cost to me if I participate?

There will be no cost to you for participating in this research study, but you will need to allow enough time for the clinic visits.

You will not be paid for being in this study; all the tests, examinations and study medication will be provided to you at no cost. However, your study doctor will ask you if you had to pay any costs (for example bus, train, taxi fares) in order to come to the clinic. You should keep all receipts of bus, train, taxi fares etc, and give them to your study doctor. You will be reimbursed for all reasonable travel expenses incurred as a result of taking part in the study.

What if something goes wrong or if I have problems while I am in the study?

If you experience a study related injury, you will be reimbursed for medical expenses for the treatment of bodily injuries that were caused by the use of the study medication.

An insurance policy that covers any damage to your health arising from your participation in this study has been taken out.

Compensation for a study related injury would be in accordance with established standards set by the Association for British Pharmaceutical Industry (ABPI). The study doctor will give you a copy of the ABPI-guidelines upon your request.

In case of study related injury please contact:

Doctor: N/A

Contact Numbers: N/A

If you are harmed by your participation in the study, you will be compensated according to the guidelines of the pharmaceutical industry relating to clinical trials (i.e. ABPI guidelines). If you are harmed due to someone's negligence then you may have grounds for legal action. You are not waiving your legal rights by signing this form.

For further information on the insurance coverage, please ask the study doctor.

Can I withdraw or be withdrawn from the study?

Taking part in the study is voluntary. If you decide to take part in the study, you are free to withdraw from the study at any time. If you decide to withdraw from the study, you should inform your study doctor immediately. Your study doctor will not be upset and you will not be penalized in any way, and your future care will not be affected. Should you withdraw from the study, the study data collected before your withdrawal may still be processed along with other data collected as part of the study.

In addition, circumstances may arise that will lead to the ending of your participation in this study. Such circumstances could include:

- Medical reasons including pregnancy;
- You not taking your medication or not regularly keeping to the study appointments, or if you do not follow the doctor's instructions;
- You taking a medication that is not allowed in this study;
- If there are not enough patients in the study;
- If the Medicines Control Council or the Committee for Pharmaceutical Trials stops or suspends the investigation.
- If the Sponsor stops the study.

Will my participation in this study be kept confidential?

The records that identify you will be kept confidential and, to the extent permitted by the applicable laws and regulations, will not be made publicly available. If you consent to take part in this study, any of the medical records and data recorded in this study may be directly inspected by representatives of the study Sponsor (x), the Medicines Control Council of South Africa and other Medicines Regulatory Authorities, the appointed staff at Tijger Trial Centre (e.g. safety monitor, etc), Pharma-Ethics and the Biomedical Research Ethics Committee of the University of the Western Cape to make sure that the study is being done correctly. By signing this written informed consent form you are giving permission for this to be done.

The information collected during the study will be stored in a computer but your name will not be stored. Only your study doctor will know that the information that is related to you. All blood collected during the study will be labeled with your anonymous subject number and initials. Samples will be sent to a central laboratory for analysis in batches, but your name will not be included on any specimens or accompanying documentation that is sent to the laboratory.

The results of the study may be published in the medical literature and/or presented at a scientific conference or symposium, but your identity will not be revealed. The information disclosed will be collective summarized data.

You may ask to see your medical information as prescribed by law. The treatment that you received in the study needs to remain unknown (blinded) until the study data is analyzed; you may see this information, but only after the data has been analyzed.



UNIVERSITY *of the*
WESTERN CAPE

PATIENT INFORMED CONSENT FORM

Patient identification number... /.....

Patient initials:

The title of the study: A randomized double-blind placebo-controlled study to investigate the efficacy of *Sutherlandia frutescens* (*subspecies microphylla*) in adult type-2 diabetics.

Protocol number: UWC 16-002 version: 04

Sponsor: N/A

By signing and dating this document,

- I confirm that I have had time to carefully read and understand the patient information sheet provided for this study.
- I confirm that I have had the opportunity to discuss the study and ask questions and I am satisfied with the answers and explanations that I have been provided.
- I give permission for any medical records to be reviewed by the sponsor or designee, and/or representative of the medicines control council and pharma ethics.
- I understand that my participation is voluntary and that I am free to withdraw at any time without giving any reason and without my medical care or legal rights being affected.
- I confirm that I have received a signed and dated copy of the patient information sheet and informed consent form.

VOLUNTEER:
Name (Capital letters) Signature Date

WITNESS:
(Where required)
Name (Capital letters) Signature Date

CLINICAL INVESTIGATOR:

.....
Name (Capital letters) Signature Date

APPENDIX 2

Documentation for MCC – Clinical trial application

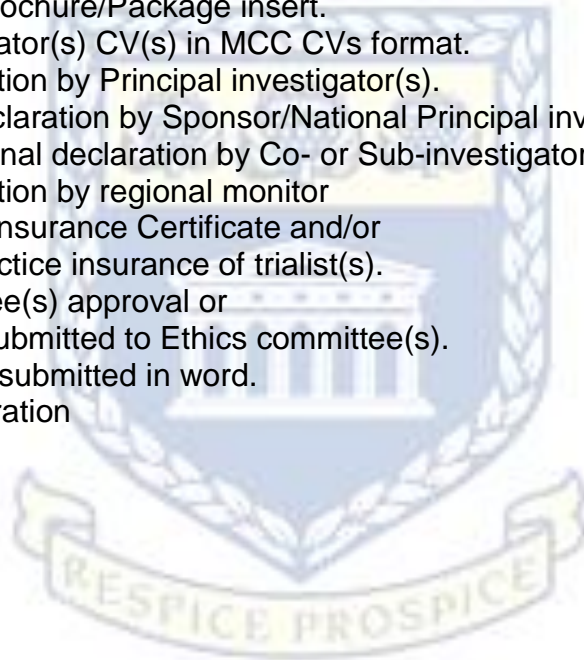


UNIVERSITY *of the*
WESTERN CAPE

APPLICATION TO CONDUCT A CLINICAL TRIAL

The following are the requirements when submitting a clinical trial application.

1. Covering letter.
2. Cover sheet.
3. Checklist.
4. Completed Application form.
5. All documents to be submitted in duplicate with two electronic copies.
6. Additional 25 copies of the application form itself must be submitted.
7. Protocol
8. Patient Information leaflet and Informed consent form
9. Standardized MCC contact details/wording to be added to PILs.
10. Investigators Brochure/Package insert.
11. Signed investigator(s) CV(s) in MCC CVs format.
12. Signed Declaration by Principal investigator(s).
13. Signed joint declaration by Sponsor/National Principal investigator.
14. Signed Provisional declaration by Co- or Sub-investigators
15. Signed Declaration by regional monitor
16. Indemnity and Insurance Certificate and/or
17. Proof of Malpractice insurance of trialist(s).
18. Ethics committee(s) approval or
19. Copy of letter submitted to Ethics committee(s).
20. Diskettes to be submitted in word.
21. Financial declaration



UNIVERSITY of the
WESTERN CAPE

SOUTH AFRICA: CLINICAL TRIAL APPLICATION

SECTION 1 – CHECK-LIST OF REQUIRED DOCUMENTATION

To be completed by Applicants for all Clinical Trials

COVER SHEET

Study Title: A randomized double blind placebo controlled study to investigate the efficacy of *Sutherlandia frutescens* (subspecies *microphylla*) in adult type-2 diabetes mellitus

Protocol No: UWC 16-002

Version No: 01

Date of Protocol: November 2015

Study Drug: *Sutherlandia frutescens*

MCC Ref number (if applicable): N/A

MCC Ref number(s) of comparator drug(s) (if applicable): N/A

MCC Ref number(s) of concomitant drug(s) (if applicable): N/A

Date(s) MCC approval of previous protocol(s): N/A

Sponsor: School of Pharmacy, University of the Western Cape

Applicant: Prof James Syce

Contact Person: Prof James Syce

Address: School of Pharmacy

University of the Western Cape

Private Bag X17, Bellville, 7535

Telephone Number: 021 959 2192

Fax Number: 021 959 1324

Cell Number: 082 202 3315

E-mail address: jsyce@uwc.ac.za

To be completed by MCC

Date original application received:

Tracking No:

Proposed Clinical Trials Committee Meeting Date if applicable:

Signature:

Date:

ACKNOWLEDGEMENT OF RECEIPT OF CTA (Contact details to be completed by the applicant). Whole cover sheet to be faxed to applicant once details in block above are completed.

Contact Details: Name:

Fax No.:

Receipt of new application is hereby acknowledged.

Date:

Signature (of MCC recipient):

Name:

CHECKLIST

Applicant

**Check list
check**

MCC

Check list (double-

- | | | |
|--------------------------|--|--------------------------|
| <input type="checkbox"/> | COVERING LETTER | <input type="checkbox"/> |
| <input type="checkbox"/> | FULLY COMPLETED APPLICATION (SECTIONS 1-3) | <input type="checkbox"/> |
| <input type="checkbox"/> | PROTOCOL (INCLUDING RELEVANT QUESTIONNAIRES ETC.) | <input type="checkbox"/> |

PATIENT INFORMATION LEAFLET(S) AND INFORMED CONSENT(S)

INVESTIGATORS BROCHURE AND / OR ALL PACKAGE INSERT(s)

INVESTIGATOR'S CV(s) IN MCC FORMAT

SIGNED DECLARATION(s) BY INVESTIGATOR(s)

REGIONAL MONITOR'S CV AND DECLARATION

CERTIFICATE(S) OF ANALYSIS *(May be submitted with ethics approval letter)*

INSURANCE CERTIFICATE AND IF NECESSARY:

LETTER ENDORSING GENERIC INSURANCE CERTIFICATE

ETHICS APPROVAL

OR

COPY OF LETTER APPLYING FOR ETHICS COMMITTEE APPROVAL

COPY/IES OF RECRUITMENT ADVERTISEMENT(s) (IF APPLICABLE)

FINANCIAL DECLARATION (SPONSOR AND NATIONAL PI)

Electronic versions of the application form (Sections 1 – 3), the Protocol, the Investigator's Brochure and/or other relevant documents:

LABELLED DISKETTE/CD-ROM (MSWORD OR RICH TEXT FORMAT)

Please List files submitted on diskette/CD-ROM:

1.....

2.....

Declaration by applicant:

We, the undersigned have submitted all requested and required documentation, and have disclosed all information which may influence the approval of this application.

We, the undersigned, agree to ensure that if the above-said clinical trial is approved, it will be conducted according to the submitted protocol and South African legal, ethical and regulatory requirements.

Applicant (local contact)

Date

National Principal Investigator /

Date

National Co-ordinator /
Other (state designation)



UNIVERSITY of the
WESTERN CAPE

SECTION 2 – ADMINISTRATIVE AND SUPPLEMENTARY DETAILS

Title: A randomized double blind placebo controlled study to investigate the efficacy of *Sutherlandia frutescens* (*subspecies microphylla*) in adult type-2 diabetes mellitus

Protocol Number/identification: UWC 16-002

Date of protocol (initial/final):

Part 1: CONTACT DETAILS (NAME/ADDRESS/TEL/CELL/FAX/E-MAIL)

1.1 Applicant: (as in Section 1)

Prof James Syce

School of Pharmacy

Dept of Pharmacology

University of the Western Cape

Private Bag X17

Bellville 7535

Tel no 021 959 2192 Fax no 021 959 1324

Cell no 082 202 3315 e-mail jsyce@uwc.ac.za

1.2 Sponsor: (as in Section 1)

School of Pharmacy

Dept of Pharmacology

University of the Western Cape

Private Bag X17

Bellville 7535

1.3 If no sponsor – person or organization initiating, managing, and / or funding the clinical trial:

- **Not applicable**

1.4 Local Contact Person for correspondence:

Prof: James Syce

School of Pharmacy

Dept of Pharmacology

University of the Western Cape

Private Bag X17

Bellville 7535

Tel no 021 959 2192

Fax no 021 959 1324

Cell no 082 202 3315

e-mail jsyce@uwc.ac.za

1.5 National Principal Investigator/Coordinator: (or equivalent person)

- **Not applicable**

1.6 International Principal Investigator: (if applicable)

- **Not applicable**

1.7 Regional Monitor: (as in Section 1)

Part 2: DETAILS OF INVESTIGATIONAL PRODUCT(S)

2.1 Name(s) and details of investigational product(s) to be used in trial:

Investigational product

Sutherlandia frutescens 400mg capsules

Placebo:

Placebo capsules which are identical in appearance to the investigational product containing a mixture of lactose, starch and small amount of dried lettuce leaf powder

[Formulation(s) and strength(s) (e.g. 10 mg/ml–10ml amp.)] *Include MCC registration number and date of registration if applicable.*

The investigational product: Sutherlandia frutescens 400mg capsules (20mm x 6mm capsules)

Placebo: identical in appearance to the investigational product containing a mixture of lactose, starch and small amount of dried lettuce leaf powder.

Dose Administration: participants will take either the investigational product or placebo in capsule form daily with food.

2.2 Name(s) and details (as above) of comparator product(s) and MCC registration number(s) and date(s) of registration if applicable:

Not applicable

2.3 Name(s) and details (as above) of concomitant medication(s) including rescue medications which are required in the protocol, and MCC registration number(s) if applicable: *[Ensure package inserts or complete pharmacological information has been included with application (Section 1)]. None*

2.4 Estimated Quantity of Trial Material (each drug detailed separately) for which exemption will be required. Not applicable

- 2.5 If any of the above drugs are available in South Africa, give an explanation for not using what is available in South Africa. Not applicable
- 2.6 Details of receiving of drugs from supplier, storage, dispensing, packaging of drugs. Not applicable
- 2.7 Date MCC registration applied for – or envisaged date of application for trial medication. Explain if registration is **not** envisaged. Trial for academic reasons only:
- Application is not for registration of the trial medication. At this stage this is a research project designed to determine the efficacy of *Sutherlandia frutescens*.
- 2.9 Registration status of entity, for the indication to be tested in this trial, in other countries: (i.e. Country: date registered / date applied for / date registration refused / date registration withdrawn by applicant / date registration cancelled by regulatory authority) [*Attach as an appendix if necessary.*].

Not registered

Part 3: DETAILS OF TRIALIST(S) AND SITE(S)

- 3.1 Details of Investigator(s): [*designation, title: (i.e. principal investigators / investigators) Include Name/Address/Tel/Cell/Fax/E-Mail*]

Principal Investigator: N/A

Investigator: *Prof James Syce*

School of Pharmacy

University of the Western Cape

Private Bag X17

Bellville 7535

Tel no: 021 959 2192

Fax no: 021 959 1324

Cell no: 082 202 3315

e-mail: jsyce@uwc.ac.za

Co-Investigator: N/A

- 3.2 Current work-load of Investigator(s): (*Number of studies currently undertaken by trialist(s) as principal and/or co- or sub-investigator, and the total number of patients represented by these studies. Time commitment of researcher(s) in relation to clinical trial work and non-trial work.*)

Recommended format for response:

Investigator (Name and designation):	Prof James Syce		
Total number of current studies (all stages) on specified date	Number 1	Date	
Total number of patients / participants for which responsible on specified date	Number 100	Date	
ESTIMATED TIME PER WEEK [168 hours denominator]	Hours	%	
Clinical trials	Clinical work (patient contact)	20	12
	Administrative work	5	3
<u>Organisation</u> (Practice / university / employer)	Clinical work	5	3
	Administrative work	5	3
Teaching	Preparation / evaluation	60	36
	Lectures / tutorials	40	24
<u>Writing up</u> work for publication / presentation		5	3
<u>Reading</u> / sourcing information (e.g. internet searches)		8	5
<u>Other</u> (specify)		20	12

3.3 Details of Site(s) (Name of site, physical address, contact details, contact person, etc.)

Campus Health Centre
University of the Western Cape
Private Bag x17
Bellville 7535
Prof James Syce
Tel no: 021 959 2192
Cell no: 082 202 3315

Fax no: 021 959 1324
e-mail: jsyce@uwc.ac.za

3.4 Capacity of Site(s): (Number of staff, names, qualifications, experience -- including study co-coordinators, site facilities, emergency facilities, other relevant infrastructure)

- 2 Full time Medical Doctors
- 3 Full time Nursing sisters trained in Primary Health Care
- 3 Full time Administrative staff.

Part 4: PARTICIPANTS (SUBJECTS)

- 4.1 Number of participants in South Africa: 100
- 4.2 Total number of participants worldwide: Not applicable
- 4.3 Total enrollment in each SA centre: *(if competitive enrollment, state minimum and maximum number per site.)*. 100 participants
- 4.4 Volunteer base from which South African participants will be drawn: Day hospitals and university campus
- 4.5 Retrospective data indicating potential of each site to recruit required number of patients within envisaged duration of trial. (SA Guidelines 2000, Item 3.3, p15) *[May be attached. Label clearly as 'Section 2 Item 4.5']*

Part 5: OTHER DETAILS

- 5.1 If the trial is to be conducted in SA and not in the host country of the applicant or sponsor, provide an explanation: Not applicable
- 5.2 Estimated duration of trial: 6 Months ± 30 days
- 5.3 Name other Regulatory Authorities to which applications to do this trial have been submitted, but approval has not yet been granted. Include date(s) of application: Not applicable
- 5.4 Name other Regulatory Authorities which have approved this trial, date(s) of approval and number of sites per country: Not applicable

5.5 If applicable, name other Regulatory Authorities or Ethics Committees which have rejected this trial and give reasons for rejection: N/A
--

- 5.6 If applicable, details of and reasons for this trial having been halted at any stage by other Regulatory Authorities: Not applicable
- 5.7 Details if this trial is being undertaken in SADC, any other country in Africa, or any country where there is no regulatory control of clinical trials: Not applicable
- 5.8 Previous studies using this agent which have been approved by MCC: none known

MCC approval number: **Not applicable**

Study title:

Protocol number:

Date of approval:

National PI / Principal Investigator:

Date(s) Progress report(s):

Date Final report:

- 1.9 If any sub studies are proposed as part of this protocol, indicate whether or not they will also be done in South Africa. If not, please explain.
Not applicable

Part 6: ETHICS

- 6.1 Ethics Committee responsible for each site, date of approval or date of application:

Pharma Ethics for both sites

- 6.2 Attach copy of response(s) made by, and/or conditions required by ethics committee(s) if available. Ensure that date of EC response is legible.

Not applicable

- 6.3 State which Good Clinical Practice (GCP) guidelines are being followed. (*Particular reference to the South African guidelines required*): SA GCP and WHO guidelines for Herbal Medicines Trials

- 6.4 Details of capacity building component of the trial, if any:

Campus Health Centre and School of Pharmacy to be developed for future clinical trials as part of South African Herbal Science and Medicines Institute at the UWC.

- 6.5 Details of the training of investigators, monitors, and study co-ordinators in terms of carrying out this trial and in terms of GCP: Attending accredited GCP courses every 2 years

- 6.6 Detailed safety and monitoring plan for each site: *[May be attached. Label as 'Section 2 Item 6.6']*

Safety issues:

An independent monitor will ensure Adverse Events are reported for both sites according accepted guidelines. Project Principle Investigator will review safety reports on an ongoing base.

- 6.7 Details of trial insurance certificate: (e.g. title, protocol, dates, policy #, amount).

The UWC currently has insurance cover, but in the event that the study will be approved and conducted, special cover will be obtained from the Insurance Company for the duration of the trial.

- 6.8 Details of possible conflict of interest of any person(s)/organisation(s) who/which will be involved in the trial:

No conflict of interest

- 6.9 Remuneration to be received in SA Rands: (Investigators) (Trial participants) (Others). Indicate broad breakdown of costs to be covered by this amount – if applicable. [Note: the CTC recommends a minimum compensation of R50.00 per visit for participants travel and incidental expenses.].

No remuneration for trial personnel, as it is for academic purpose only. Study subjects will receive R50.00 travel allowance.

Reviewer's comments on Section 2:

SECTION 3 – APPLICANT'S REPORT / PRESENTATION

[Please use Black 12 point Arial Font, using MSWord or rich text format (rtf) for electronic version]

1. Title: A randomized double blind placebo controlled study to investigate the efficacy of *Sutherlandia frutescens* (subspecies *microphylla*) in adult type-2 diabetes mellitus

CTC Reviewer's comment:

2. Protocol Number/identification: UWC 16-002
3. Rationale for study summarised: (Why should this trial be done at all?). Include statement about South African contribution, if any, to the development of this protocol.

Type-2 diabetes mellitus is the most common form of diabetes, accounting for 90 - 95% of cases (Centers for Disease Control, 2011). It has been further estimated that diabetes prevalence for South Africans is at 6.46% for adults aged between 20-79 years. *S. frutescens* is thought to be among the most efficacious plants used in Southern African traditional medicine. The plant has been used in indigenous settings to treat diabetes, as well as inflammatory conditions. It's possible that *S. frutescens* may have useful effects in adult's type-2 diabetes.

There has been few properly conducted clinical trials performed on traditionally used herbal medicines in South Africa and *S. frutescens* is no exception. Nevertheless, this

herbal medicine is being used widely without any apparent recorded adverse effects and has the potential to be of immense benefit.

Given the widespread indigenous use of *S. frutescens* as a phytotherapeutic and its non-toxic effects are shown in the pre-clinical studies in a primate, it is logical to assess its efficacy, during chronic use, in clinical trials.

In both the traditional and commercial settings, the normal recommended dose of *S. frutescens* is approximately 600 to 800 mg daily. However, it is known that in the traditional setting doses are substantially higher than this frequently used and/or the *S. frutescens* combined with other plant medicines. The outcomes of the completed phase I study showed that 800mg (12 mg/kg for an average 67 kg human) daily doses of *S. frutescens* leave powder produced no serious effect while doses of 3x and 9x (i.e. 81 mg/kg body weight) the equivalent human dose also produced no adverse effects in the vervet monkey. To optimize the chance of clearly establishing whether *S. frutescens* is safe and efficacious or not in the traditional setting, a dose-escalating study to determine the bioavailability for the safe and effective use of the product would thus be well justified.

CTC Reviewer's comment:

4. Background information (**summarised** – **essential** points that apply to this) [1-2 sentences max for each point]:

Disease / problem: Type-2 diabetes mellitus
South African context (e.g. local epidemiology)
Properties of Drug / Entity; hypotheses about mechanism of action, etc.

Sutherlandia is thought to be among the most efficacious plants used in southern African traditional medicine. The medicinal value of *S. frutescens* has been ascribed to a variety of its constituents including pinitol, γ -amino butyric acid (GABA), and l-canavanine.

Pre-clinical findings: (e.g. laboratory / animal / toxicity / mutagenicity)

A recently completed study on vervet monkeys (*Chlorocebus aethiops*) conducted by the South African Medical Research Council (MRC), found that up to nine recommended dose of *S. frutescens* (81 mg/kg body weight per day for 3 months) resulted in no significant changes to relevant haematological, biochemical and physiological parameters.

Clinical findings (e.g. phases; PK; PD; dose-finding; ADRs, NNT/NNH, other)

Systematic review(s) and/or citations per year-group on a Medline search

Results of Phase I of a double-blind placebo-controlled study conducted by The International Center for Indigenous Phytotherapy Studies through the South African Herbal Science and Medicine Institute, University of the Western Cape have just been made available (Johnson, Syce, Nell, Rudeen, & Folk, 2007). In this double-blind placebo randomized controlled study, the safety of *L.*

frutescens in 25 healthy adult volunteers was evaluated. Twelve healthy subjects were selected for the treatment arm and received 400 mg *L. frutescens* leaf powder capsules twice a day. In the control arm, 13 subjects received an identical placebo capsule twice a day. Participants were seen at monthly intervals throughout the 3-month study period. Blood and urine samples for haematology/biochemistry (i.e., Red Blood Cell (RBC), White Blood Cell (WBC), serum analysis and serum proteins) and urinalysis (i.e., Serum Glutamate (SG), Potential of hydrogen (pH), protein, glucose, ketone, bilirubin, blood, & urobilinogen) were measured. Participants also completed a 12-lead Electrocardiogram (ECG) and reported any adverse events at each visit. Results indicate that the 800 mg daily dosage was well tolerated by study participants, with no significant changes present in the relevant haematologic, biochemical or physiological parameters (Johnson *et al.*, 2007).

CTC Reviewer's comment:

5. Objectives of study (clearly listed and justified)

- To determine the efficacy of *Sutherlandia frutescens* (subsp. *Microphylla*) capsules in adult type-2 diabetes mellitus by assessing, HbA1C.
- To evaluate the proportion of participants achieving a therapeutic response (HbA1c < 7%) with *S.f* 1200mg versus placebo after 12 weeks of treatment
- To evaluate the effect of *Sutherlandia frutescens* on glycemic control (HbA1c, fasting plasma glucose, etc).

CTC Reviewer's comment:

6. Study design (clearly described and each component justified) [includes phase, use of placebo, dosages, randomisation, blinding, duration, etc.]

A 6-month a randomized double blind placebo controlled study. Participants will be screened at Visit 1 and eligible subjects will, at visit 2, be randomized to either *Sutherlandia frutescens* (subsp. *Microphylla*) or placebo. 1200mg capsule, b.i.d or a placebo a similar appearance.

CTC Reviewer's comment:

7. Participants: (number of participants; ability to enroll required number within stated time)

Total participants: 100

CTC Reviewer's comment:

8. Eligibility and enrollment: (Inclusion and exclusion criteria listed and justified)

CTC Reviewer's comment:

Inclusion criteria

9. T2DM man and woman between 18-70 years of age on a stable medication.
10. HBA1C >6.5% <8.5%
11. Fasting plasma glucose (FPG) 7.0-14.0 mmol/l
12. Have body weights within 25% of the appropriate weight range
13. Had taken standardized diet control + exercise therapy + stable administration of metformin for more than 3 months before being enrolled.
14. Has a normal ECG as determined by a unit physician.
15. Has not taken any traditional medication for 28 days prior to screening.
16. Be informed of the nature of the study and will give written informed consent.

Exclusion criteria

17. Type 1 diabetes mellitus, gestational diabetes mellitus and other special types of diabetes mellitus.
 18. In the past 3 months, had taken medication to control body weight (including weight-loss drug) or oral administration of any anti-diabetic drugs or insulin except for metformin.
 19. Had diabetic ketosis, ketoacidosis.
 20. Fasting triglycerides >450mg/dL (>5.1 mmol/L)
 21. Had uncontrolled hypertension (blood pressure be and more than 160/100 mmHG).
 22. Intake of anti-diabetic medication within 14 days before the start of the study.
 23. Had mental illness, alcohol addiction and /or taken administration of psychoactive drug substance.
 24. Smokers, who smoke more than 10 cigarettes per day and cannot, refrain from smoking during the study period.
 25. Pregnancy, lactation or being prepared pregnant woman
 26. Had allergic constitution.
 27. Unwillingness or inability to follow the procedures outlined in the protocol
 28. Any disease or condition which might compromise the haematopoietic, renal, endocrine, pulmonary, central nervous, cardiovascular systems.
9. Treatment modalities and regimens, drug accountability [clearly explained and justified for all participant groups/arms e.g. in terms of route of administration, dose, etc. Drug accountability clearly described.]

Investigational product

Sutherlandia frutescens 400mg capsules

Placebo:

Placebo capsules which are identical in appearance to the investigational product containing a mixture of lactose, starch and small amount of dried lettuce leaf powder

[Formulation(s) and strength(s) (e.g. 10 mg/ml–10ml amp.)] *Include MCC registration number and date of registration if applicable.*

The investigational product: Sutherlandia frutescens 400mg capsules (20mm x 6mm capsules)

Placebo: identical in appearance to the investigational product containing a mixture of lactose, starch and small amount of dried lettuce leaf powder.

Dose Administration: participants will take either the investigational product or placebo in capsule form daily with food.

CTC Reviewer's comment:

10. Outcome measurements/variables *(each clearly stated and justified)*

Primary Safety

The following primary safety endpoints will be measured at each study visit from the beginning of screening to week 24.

Adverse events will be measured at all study visits following the start of the study medication.

Secondary Safety Endpoints

The following secondary endpoint will be measured at baseline, week 4, week12 and week24.

Efficacy Endpoints

The following efficacy endpoint HbA1c will be measured at week 24.

CTC Reviewer's comment:

11. Adverse events *(prevention, definitions – including causality assignment, recording, reporting, time-lines, action to be taken, all clearly described).*

An adverse event is the development of an undesirable medical condition - e.g. symptoms or abnormal results of an investigation - or the deterioration of a pre-existing medical condition (not relevant in this study). AE's will be collected by means of a standard question: "Have you had any health problems since the previous visit?" AE's will be recorded at every visit. Spontaneously reported AE's and/or observed AE's and the subject's response to this question will be recorded on the AE form with information about seriousness, the action was taken, date of onset and recovery, maximum intensity, and outcome. The subjects will be asked to assess the intensity of the reported Adverse Event according to the following scale:

Mild = awareness of sign or symptom, but easily tolerated

Moderate = discomfort sufficient to cause interference with normal activities

Severe = incapacitating, with the inability to perform normal activities.

A Serious Adverse Event is an adverse event occurring during any phase of the study and at any dose of the investigational product or placebo, which fulfils one or more of the following criteria:

- Results in death;
- Is immediately life-threatening;
- Subject hospitalization;
- Results in persistent or significant disability or incapacity.

The causality of Serious Adverse Events (i.e. the relationship to study treatment) will be assessed by the investigators, who in completing the relevant Case Report Form must answer 'yes' or 'no' to the question "Do you consider that there is a reasonable possibility that the event may have been caused by the study medication?" The following factors should be considered when deciding if there is a "reasonable possibility" that an Adverse Event may have been caused by the investigational product.

- Time course of events and exposure to suspect drug – did the AE occur in a reasonable temporal relationship to the administration of suspect drug?
- De-challenge experience – did the AEs resolve or improve on stopping or reducing the dose of the suspect drug?
- Re-challenge experience - did the AEs reoccur if the suspected drug was reintroduced after having stopped?
- Laboratory tests – has a specific laboratory investigation confirmed the relationship?
- No alternative cause, the AEs cannot be reasonably explained by aetiology such as an underlying disease (not previously present), other drugs or environmental factors.

There would not be a "reasonable possibility" of causality if none of the above criteria apply or where there is evidence of exposure and a reasonable time course, but any de-challenge is negative or there is another more likely cause of the AEs.

In this study, the Adverse Events will be noted from the interview at the time of visit and from the daily input in the diary.

CTC Reviewer's comment:

12. Statistical measures:

- Determination of sample size correct, clear and justified (with and/or without stratification)
- Statistical method(s) and analysis of quantitative measures appropriate, clear and justified
- Statistical method(s) and analysis of qualitative measures appropriate, clear and justified
- Data processing (how, where, when, who) clearly described and justified. If a SA person will be involved in data processing, please give details.

- Interim analysis envisaged or not (justify) and stopping rules if applicable (explain)

CTC Reviewer's comment:

13 **Ethical Issues:** justification of 'Section 2 part 6' including:

- Explanation of which GCP guidelines are or are not being followed – with particular reference to the South African guidelines
- Comment on choice of investigators (*refer to point C of Introduction, page 2 SA Clinical Trials Guidelines 2000*)
- Comment on need for, appropriateness of, and relevance of GCP training / updating / for staff involved in this trial
- Comment on capacity building element of trial
- Comment on resources of sites and sponsor
- Comment on monitors and monitoring plan
- Indicate how additional staff (monitors, pharmacists, nursing staff, etc.) will maintain patient confidentiality, follow the protocol, and abide by ethical and regulatory requirements
- Comment on insurance and indemnity measures
- Comment on Patient Information Leaflet and Informed Consent (NB: inclusion of ABPI guidelines; appropriate level of education/English; possible benefits / risks clear; ensuring patient rights; contact names and numbers, as well as MCC details, included)
- Comment on availability and completeness of separate PILs and informed consent forms for any proposed archiving of blood specimens for later research or for genetics research.
- Comment on ethics of the publication policy
- Comment on treatment and/or management of participants and their disease condition(s) after completion of trial
- Comment on ethics committee capacity to monitor site if not a local ethics committee
- Provide an explanation if minimum recommended compensation for participants is not being provided.

CTC Reviewer's comment:

14. Other relevant information not included above

- *e.g. Are references adequate and dates of references current?*
- *Are there discrepancies between protocol and IB or package inserts? Are there specific explanation(s) for these discrepancies?*
- *Are the explanations for not following the SA 'GCP guidelines' acceptable?*
- *Any other comments on this trial.*

None

CTC Reviewer's comment:

For office use

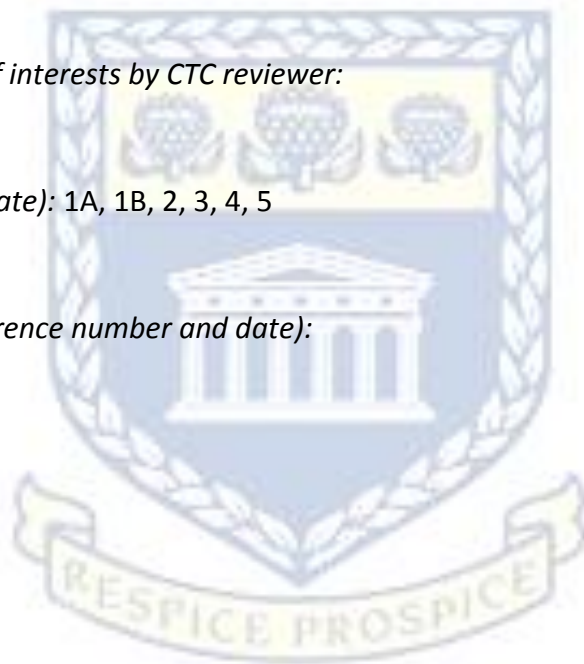
CTC Reviewer's questions and concerns to be considered and/or forwarded to applicant:

CTC Reviewer's recommendation:

Declaration of conflict of interests by CTC reviewer:

CTC recommendation (date): 1A, 1B, 2, 3, 4, 5

MCC decision (MCC reference number and date):



UNIVERSITY of the
WESTERN CAPE

Appendix 3A

SUMMARY OF REQUIREMENTS FOR ETHICS APPLICATIONS SUBMITTED TO STELLENBOSCH UNIVERSITY HREC



1. SUBMISSION OF AN ETHICS APPLICATION FOR *CLINICAL TRIALS*

1.	2 hard copies of full application
	Submit to Elvira Rohland, room 5008, 5th floor, teaching block
2.	1 electronic copy of full application
	<ul style="list-style-type: none">• Submit in one email to <u>ethics@sun.ac.za</u>• Submit the protocol and any other documents created in Microsoft word as either word documents or .pdf files• Submit supporting documents as individual .pdf files <i>e.g. one .pdf file for the application form, one .pdf file for the checklist, one .pdf file for each declaration letter, one .pdf file for each CV, etc.</i>• Submit a scanned .pdf file of each signed document

2. SUBMISSION OF AN ETHICS APPLICATION FOR *HUMAN AND STUDENT RESEARCH*

1.	1 hard copy of full application
	Submit to Elvira Rohland, room 5008, 5th floor, teaching block
2.	1 electronic copy of full application
	<ul style="list-style-type: none">• Submit in one email to ethics@sun.ac.za• Submit the protocol and any other documents created in Microsoft word as either word documents or .pdf files• Submit supporting documents as individual .pdf files <i>e.g. one .pdf file for the application form, one .pdf file for the checklist, one .pdf file for each declaration letter, one .pdf file for each CV, etc.</i>• Submit a scanned .pdf file of each signed document

3. *Clinical trial ethics application Stellenbosch University (Protocol synopsis)*

Submit a 2-page protocol synopsis or summary of the proposed research, in addition to the full protocol.

Include the following:

1. Title.
2. A short introduction, motivation and literature overview (1 paragraph only).
3. Research question or hypothesis.
4. Aims and objectives.
5. A concise summary of the methodology.
6. Description of subject population including characteristics, age range and number of subjects.
7. If the research will require blood samples, bone marrow biopsy samples, other biopsies or the collection of tissues, etc., performed solely because of participation in the research, please indicate the exact amounts and frequency with which the samples will be taken.
8. Anticipated risks as well as the precautions taken to minimize risk.
9. Anticipated benefits.
10. Ethical Considerations.

2. *Participant information and consent form (PICF)*

2. Submit your Informed Consent Form(s) to our HREC in either Afrikaans or English.

3. HREC may request changes for re-submission.
4. After approval of the original consent form, submit translations in English, Afrikaans, and Xhosa, along with a translation certificate or letter of authenticity.

3. *Short curriculum vitae of ALL investigators*

Submit a short CV for the principal investigator, co-investigators, and sub-investigators.

Each CV should not comprise more than 2 pages.

4. *Investigator declaration for ALL investigators*

Complete and sign "investigator declaration" and declare any conflict of interest for the principal investigator, co-investigators, and sub-investigators.

If the study is for degree purposes, a supervisor declaration should be signed by the study supervisor.

Investigator/Supervisor Declaration.

5. *Submission of research protocols*

Additional documentation that must accompany clinical trial applications:

1. Cover letter.
2. Flowchart.
3. A description of the study site, including the available infrastructure and the roles and responsibilities of study staff.
4. MCC approval or proof of application (if applicable).
5. NHREC approval or proof of application.
6. Proof of insurance for participants (if applicable).
7. Material for distribution to patients, including diary cards, QOL questionnaires, etc.
8. Recruitment material and advertisements (if applicable).
9. Proof of GCP training.
10. SA approved package insert(s) of registered comparators.
11. Investigator's brochure (if applicable).
12. Payment instruction forms (if applicable).

All documentation should be properly indexed, with the protocol number clearly visible on all the sections of each document. All relevant attachments with regard to the study staff should be put together per person, i.e. CV, Declaration, Health Professions Council of South Africa (HPCSA) / *South African Nursing Council (SANC)* Registration, Malpractice Insurance, GCP Certificate, Dispensing Licence, etc. The manual and computer filing systems of the REC are based on the protocol number and not the name of the drug involved.

Covering Letter: The covering letter must give a brief summary of the protocol and indicate the study doctor's assessment of any potential additional risk or discomfort to the participants. Only the names of investigators/sub-investigators and all study staff in the private healthcare sector, on behalf of whom the application is made, must be listed in the letter with an indication of their submitted documents.

Language: The Committee will only consider and approve English documentation. South African English spelling should be used in all documents, including the Patient Information Documents. Should translations be required, the sponsor or investigator(s) (in non-sponsor driven research), must obtain the services of a professional translator, and keep a record of their certification as to the accuracy of the translation. Where research involves the participation of persons unfamiliar with the language in which the research is to be conducted the Performance in Initiating and Delivering (PID) and related documentation must be translated into the participants' language.

When utilising the services of an interpreter, the investigator must ensure that the participant's informed consent is obtained and that an interpreter is present during discussions with the participants about the research study. As a rule, the interpreter should be an independent person and the patient should consent to the presence of the interpreter.

Medicines Control Council (MCC): Approval where MCC approval for the trial is required, a copy of the approval letter must be submitted. If MCC approval is pending, proof of application to the MCC must be included. Where only MCC notification is required, a copy of the notification must be submitted.

Amendments: They must be submitted to and will be approved by the Chairperson, Vice-chairperson or the REC Amendments Subcommittee, unless otherwise indicated. Such approvals will also be ratified by the full committee at the subsequent meeting. Covering letters accompanying amended PIDs must state the date of their original approval. Amendments must be shown on the latest REC approved document containing the changes recommended by REC, and the changes should be highlighted to facilitate review. All amendments must be submitted electronically.

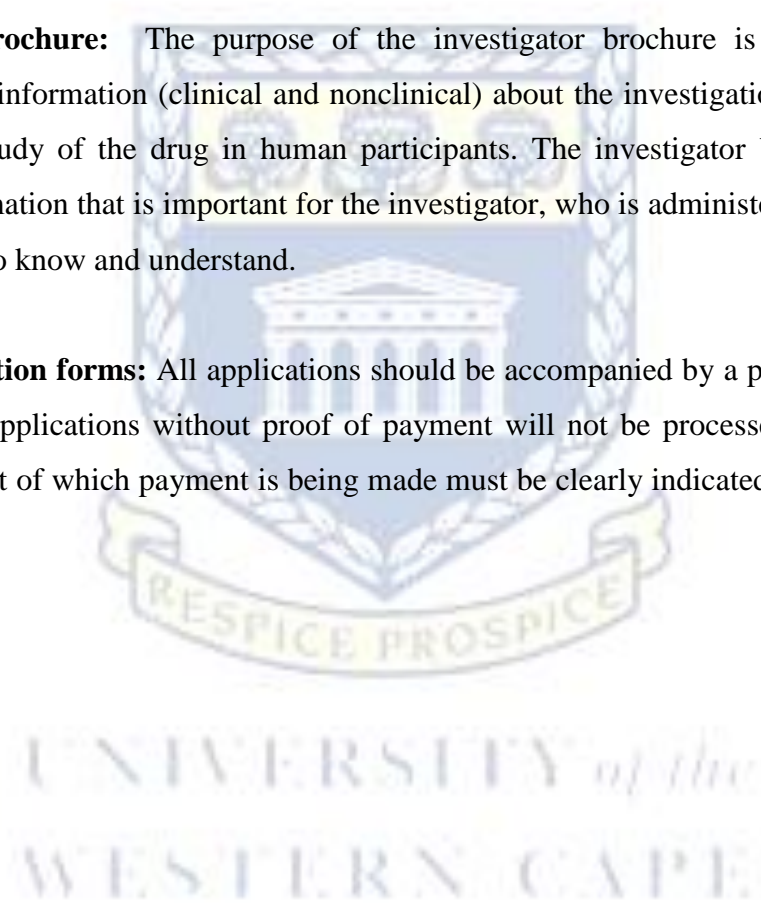
NHREC approval or proof of application: Sponsors of clinical trials must register all South African-based trials on the South Africa National Clinical Trials Register (SANCTR) which is managed by the department of health.

Proof of GCP training: Approval of GCP course training must be included of the study staff.

DECLARATIONS: Declarations by all study staff, must reflect the name of the sponsoring company, protocol number and title, as well as the study staff's name and designation. Declarations must be properly completed and signed.

Investigator's brochure: The purpose of the investigator brochure is to provide the investigator with information (clinical and nonclinical) about the investigational drug that is relevant to the study of the drug in human participants. The investigator brochure should include the information that is important for the investigator, who is administering the drug to human subjects, to know and understand.

Payment instruction forms: All applications should be accompanied by a proof of payment on submission. Applications without proof of payment will not be processed. The specific protocol in respect of which payment is being made must be clearly indicated on the proof of payment.



Appendix 3B

Documentation for Stellenbosch Institutional Review Board –

Application for registration of a research project



UNIVERSITY *of the*
WESTERN CAPE

SECTION 1: DETAILS OF APPLICANT/PRINCIPAL INVESTIGATOR				
Title, First name, Surname: Prof James Syce		SU number:	PROJECT ID NUMBER	
Professional Status:				
University DIVISION:				
University DEPARTMENT: School of Pharmacy				
Complete Postal Address: University of the Western Cape Private Bag X17, Bellville, 7535				
Telephone No: 021 959 2192		E-mail address: jsyce@uwc.ac.za		
Registration with Professional Licensing Body* (e.g. HPCSA, Nursing Council, AHPCSA) <input type="checkbox"/> Yes <input checked="" type="checkbox"/> No		Registration #:		
<p>*Note:</p> <ul style="list-style-type: none"> • or equivalent statutory health council registration no. as appropriate • if registration is pending, submit proof of application • if a non-medically trained PI is overseeing research which involves medical procedures, the application must include a medical doctor registered with the HPCSA as a co-investigator 				
SECTION 2: TITLE OF STUDY				
Title of Research Project: A randomized double blind placebo controlled study to investigate the efficacy of <i>Sutherlandia frutescens</i> (subspecies <i>microphylla</i>) in adult type-2 diabetes mellitus				
Sponsor's Protocol No (if applicable) N/A				
Sponsor's Details (if applicable) N/A				
Is this a sub-study (new research question) linked to an existing/main study? <input type="checkbox"/> Yes <input type="checkbox"/> No			If yes, HREC #:	
SECTION 3: STUDY FOR DEGREE PURPOSES (including electives and skripsies)		Yes	Undergraduate	Postgraduate ×
Name of Degree: MSc pharmaceutical science		Supervisor: Prof James Syce		
Division:		Contact No:		
Department: School of pharmacy		E-mail: jsyce@uwc.ac.za		
Is this a group student project? (if yes, please list names of all students in group under Section 4) No				
Will this project involve students as part of the research team (but <u>not</u> for degree purposes)? <input type="checkbox"/> Yes <input checked="" type="checkbox"/> No (if yes, please list names under Section 4)				

Will this project involve students as part of the research team (but not for degree purposes)? Yes No

(if yes, please list names under Section 4)

SECTION 4: DETAILS OF COLLABORATING INVESTIGATORS

Name and Title	Position and role	If investigator is a student, please indicate whether postgraduate or undergraduate	Division AND Department
1.			
2.			
3.			
4.			
5.			

SECTION 5: DETAILS OF SUB-INVESTIGATORS

Name and Title	Position and role	If investigator is a student, please indicate whether postgraduate or undergraduate	Division AND Department
1.			
2.			
3.			
4.			
5.			

SECTION 6: WHERE WILL THE STUDY BE CONDUCTED?

1. Tygerberg Hospital	
2. Stikland Hospital	
3. Karl Bremer Hospital	
4. Faculty of Medicine and Health Sciences	

5. Other: please list Clinical Research Centre, the University of Cape Town, L51 Old Main Building, Groote Schuur Hospital, Observatory Road.	
---	--

SECTION 7: HUMAN SUBJECTS RESEARCH PROTECTION	
1. Does the Research involve Human Subjects who are Alive?	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No
Dead (includes identifiable tissues specimens)?	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
Medical records only?	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
Students, staff or alumni of Stellenbosch University	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
2. Will any medicine be tested during the investigation?	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
2.1 If Yes to question 2, is the medicine approved by the Medicines Control Council?	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
2.2 If yes to question 2.1, is the medicine registered for the dose which will be used in this specific project?	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
2.3 If Yes to question 2.1, is the medicine registered for the indication(s) which will be used in this specific project?	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
2.4 If No to question 2.1, is the medicine approved by the Medicines Control Council for your use in this specific project?	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
2.5 If No to question 2.2 and/or 2.3, is the medicine approved by the Medicines Control Council for your use in this specific project?	<input type="checkbox"/> Yes <input type="checkbox"/> No
3. Will any radioactive material be administered to the patient during the investigation?	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
4. Is any biohazardous material (*) involved in the project? <i>(*) "Biohazardous material" refers to recombinant DNA molecules, viruses, fungi, parasites, bacteria and all other potentially biohazardous material or products that are dangerous to both the experimental patient and the researcher.</i>	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No

UNIVERSITY of the
WESTERN CAPE

SECTION 8: STUDY TYPE			
1. Industry Sponsored Clinical Trial	x	2. Self Initiated Clinical Trial	
3. Retrospective Record Review		4. Laboratory-Based Research	
5. Qualitative Research		6. Prospective Descriptive Study	
7. Other		Please state type if 'Other':	
SECTION 9: HOW IS THIS RESEARCH FUNDED? <i>(State approximate total budget)</i>			
1. Industry	R	2. NIH/US government funded research	R
3. Other international grant funded research (e.g. Wellcome Trust)	R	4. National grant funded research (e.g. NRF, MRC, CSIR, etc)	R
5. Harry Crossley funded research	R	6. Research funded solely from SU departmental budget	R
7. Self funded research	R	8. Non-sponsored student research for degree purposes at Stellenbosch University	R



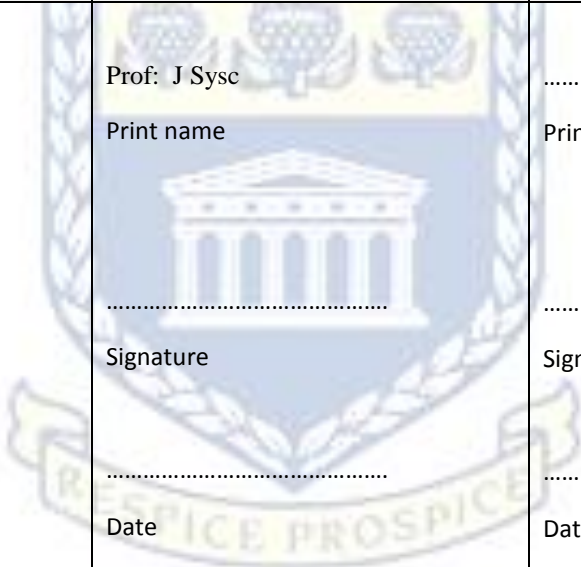
SECTION 10: RESEARCH WITH CHILDREN	
1. Does your research involve children? (A child is defined as a person younger than 18 years old)	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
<i>If no, please continue to section 9</i>	
<i>If yes, please specify the age range of potential child participants</i>	
1.1 This research is essential research for children and presents a reasonable opportunity to further the understanding, prevention, or alleviation of a serious problem affecting the health or welfare of children.	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
1.2 Indicate which risk category is applicable to your research involving children (Please check [✓] the appropriate box below and provide a brief justification)	
1.2.1 <i>The research poses no more than minimal risk to the child (that is, the risk commensurate with daily life or routine medical or psychological examinations – referred to as ‘negligible risk’ in some guidelines);</i>	
1.2.2 <i>The research poses more than minimal risk but holds out the prospect of direct benefit for the child participant.</i>	
1.2.3 <i>The research poses a minor increase over minimal risk, with no prospect of direct benefit to the child participant, but will likely yield generalisable knowledge about the condition under study;</i>	
1.2.4 <i>The research does not meet the conditions for the risk categories above but presents a reasonable opportunity to further the understanding, prevention, or alleviation of a serious problem affecting the health or welfare of children.</i>	
1.2.5 <i>Brief justification:</i>	
1.3 Indicate whether the child research is <i>Therapeutic</i> or <i>Non-therapeutic</i> (Please check [✓] the appropriate box below and provide a brief justification)	
1.3.1 Therapeutic research = Interventions that hold out the prospect of direct health-related benefit for the child participant; OR	
1.3.2 Non-therapeutic research = Interventions that do not hold out the prospect of direct health-related benefit for the child participant but results may be produced that significantly contribute to generalisable knowledge about the child participant’s condition. (If you marked “yes” to this question please ensure to complete section 1.3 below)	
1.3.3 <i>Brief justification:</i>	
1.4 Department of Health regulations for <u>non-therapeutic</u> research with children (complete only if you ticked 1.3.2 above)	
1.4.1 Condition 1: The research objectives cannot be achieved except by the participation of minors	<input type="checkbox"/> Yes <input type="checkbox"/> No
<i>Describe the scientific justification for the enrolment of minors. Explain why this research must be done with minors as participants:</i>	
1.4.2 Condition 2: The research is likely lead to an improved scientific understanding of certain	<input type="checkbox"/> Yes <input type="checkbox"/> No

conditions, diseases or disorders affecting minors	
<i>Describe how the research might, or aims to, advance knowledge affecting the health and welfare of minors as a class. Note that 'condition' is defined in the Regulations as 'physical and psycho- social characteristics understood to affect health' allowing that this research does not only involve children with an illness:</i>	
1.4.3 Condition 3: Any consent given to the research is in line with public policy	<input type="checkbox"/> Yes <input type="checkbox"/> No
<i>Consent given by authorised persons must be in line with public policy considerations. Describe how consent to the research will be in line with public policy or would be acceptable, for example, show how the research poses acceptable risks and promotes the rights of minors:</i>	
1.4.4 Condition 4: The research does not pose a significant risk to minors; and if there is some risk, the benefit of the research outweighs the risk.	<input type="checkbox"/> Yes <input type="checkbox"/> No
<i>Describe how the potential risks from the research procedures and/or intervention to minor participants will be minimized and describe any possible benefits from the research to society in the form of knowledge:</i>	
1.5 Paediatric blood volumes	
1.5.1 Please indicate the volume of blood you plan to draw from each child. _____ (including routine blood specimens for clinical care)	
<i>Please see: http://www.sun.ac.za/english/faculty/healthsciences/rdsd/Pages/Ethics/SOP.aspx for guidance on ethically acceptable blood volumes</i>	
1.5.2 If the blood volume exceeds the above guideline, please provide additional motivation for consideration by HREC:	

WESTERN CAPE

SECTION 11: DISCLOSURES	
1. Have you acquainted yourself with the code of conduct regarding the Ethics of research at this Institution and do you undertake to fully comply with it at all times?	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No
2. Has this study been, or is it likely to be, submitted to any other Research Ethics Committee?	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No
2.1 If yes, please name the Committee(s) and provide outcome i.e. approved/rejected. (If approved, attach approval letter). University of the wester cape committee	
3. Has the Principal investigator or any of the co-investigators been previously/or are presently being investigated for alleged research misconduct?	<input type="checkbox"/> Yes <input type="checkbox"/> No
3.1 If yes, please provide details and dates	
4. Are any of your intended research participants in other research studies and/or trials?	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
4.1 If yes, please provide details	
5. Are you presently a Principal Investigator (PI) in other research and/or clinical trial activities?	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
5.1 If yes, please provide details and % of your time allocated to each	
6. Have you completed a Payment instruction form: Health/Human or Payment instruction form: Clinical trial AND attached proof of payment to this application (Health/Human research)?	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No
7. Does this protocol comply with the Helsinki Declaration of 2013? (See http://www.wma.net/en/30publications/10policies/b3/)	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No
7.1 If no, please explain with full justification	
8. Does the protocol provide insurance for research-related adverse events?	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No
8.1 If yes, please describe: The insurance will be arrangement from uwc, department of pharmacolog, school of pharmacy	
8.2 If no, please justify:	
8.3 Is the provision of insurance compliant with SAGCP Section 4.11? (See Section 9 "Participant Insurance" of Health Research Ethics (HREC) Standard Operating Procedures (SOP) – Available at: http://www.sun.ac.za/english/faculty/healthsciences/rdsd/Pages/Ethics/SOP.aspx)	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No
8.4 If no, please justify:	
9. If you anticipate exporting samples/data to other site(s), locally or internationally, please provide a justification for this. Note: Attach draft Material Transfer Agreement (MTA).	
10. Does the protocol provide for payment of research participants according to National Health Research Ethics (NHREC) guidance? (See NHREC (2012). Payment of trial participants in South Africa: Ethical considerations for Research Ethics Committees (RECs). NHREC) – Available at: http://www.sun.ac.za/english/faculty/healthsciences/rdsd/Pages/Ethics/SOP.aspx	Yes R90 transport
11. Does the project involve the use of diagnostic test results (e.g. those obtained by imaging or by laboratory testing)?	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No

11.1 If yes, has the applicant consulted a professional from a relevant diagnostic discipline (e.g. radiology or pathology, as applicable)?	✓ Yes <input type="checkbox"/> No
11.2 Please provide the name, position, and discipline of person consulted: Not applicable	

SECTION 12: SIGNING OF APPLICATION		
Applicant	Supervisor <i>(only for student research)</i>	Head of Division
Mr: Ramadan Swead Print name Signature Date	 Prof: J Sysc Print name Signature Date Print name Signature Date

UNIVERSITY of the
WESTERN CAPE

DOCUMENTS REQUIRED FOR ALL SUBMISSIONS	
1. HREC application form	
2. Completed checklist	Complete either the General or Clinical Trial Checklist, whichever is applicable.
3. Payment instruction form (human/health research or Clinical Trial) AND Proof of payment (human/health research) (Non-sponsored student research for degree purposes and research funded solely from departmental budgets are exempt)	
4. Study Protocol	
5. Protocol synopsis or summary	<p>Please provide a protocol synopsis or summary of the proposed research, in addition to the full protocol, no longer than 2 pages. The Protocol Synopsis or summary should contain the following:</p> <ul style="list-style-type: none"> ▪ Title ▪ A short introduction, motivation and literature overview (1 paragraph only) ▪ Research question or hypothesis ▪ Aims and Objectives ▪ A concise summary of the methodology ▪ Description of subject population including characteristics, age range and number of subjects ▪ If the research will require blood draws, bone marrow biopsy samples, other biopsies or the collection of tissues, etc., performed solely because of participation in the research, please indicate the exact amounts and frequency with which the samples will be taken. ▪ Anticipated risks as well as the precautions taken to minimize risk ▪ Anticipated benefits ▪ Ethical Considerations
6. Participant Information and Consent Form (ICF)	<p>The ICF can be submitted in either English or Afrikaans. Once the requested changes, if any, have been made, then the HREC requests the researcher to submit translations in English, Afrikaans and Xhosa, along with a translation certificate or letter of authenticity.</p> <p><i>Note: if it has been decided that translated consent forms are not necessary for the particular study, then the applicant is required to specifically justify this in the protocol under "Ethical considerations."</i></p>
7. Short Curriculum Vitae (CV) of all investigators	Submit a short CV for the principal investigator, co-investigators, and sub-investigators. Each CV should not comprise more than 2 pages.
8. Investigator Declaration for all investigators	Complete and sign and "investigator declaration" and declare any conflict of interest for the principal investigator, co-investigators, and sub-investigators. If the study is for degree purposes, a supervisor declaration should be signed by the study supervisor.
9. Budget & Financial contract	Submit a budget (if not included in the protocol) and financial contract (if applicable i.e. external funding)
10. Draft Material Transfer Agreement (MTA), if relevant.	
ADDITIONAL DOCUMENTS REQUIRED FOR CLINICAL TRIAL SUBMISSIONS ONLY	
<p>If you are submitting a clinical trial application, please see the list below for <i>additional</i> documentation that must accompany clinical trial applications:</p> <ol style="list-style-type: none"> 1. Cover letter 2. Flow chart 3. A description of the study site, including the available infrastructure and the roles and responsibilities of study staff 4. MCC approval or proof of application (if applicable) 5. NHREC approval or proof of application 6. Proof of insurance for participants (if applicable) 7. Letter of legal indemnity, extended to Stellenbosch University and Tygerberg/ Stikland Hospital (if applicable) 8. Material for distribution to patients, including diary cards, QOL questionnaires etc. 9. Recruitment material and advertisements 10. Proof of GCP training 11. SA approved package insert(s) of registered comparators 12. Investigator's brochure 13. Payment instruction form 	