The Effect of Ginseng (The Genus Panax) on Glycemic and Vascular Health

by

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A thesis submitted in conformity with the requirements

for the degree of Master of Science

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ABSTRACT

The objective was to conduct a systematic review and meta-analysis of RCTs to assess ginseng’s effect on glycemic control and further undertake a RCT to explore the therapeutic potential of the economically-grown, non-steamed Korean ginseng variety, Korean white ginseng (KWG). The meta-analysis of 16-trials (n=770) showed that ginseng significantly improved fasting blood glucose relative to control (P=0.03), with no effect on other glycemic parameters. The RCT in 25-subjects with type-2-diabetes revealed that acute administrations of 1g, 3g, and 6gKWG were safe, but did not improve postprandial glycemia and blood pressure measures relative to control. Nevertheless, 3gKWG demonstrated significant improvements in augmentation index compared to control (P=0.04). Overall, the meta-analysis results indicate that ginseng may serve as an adjunct to conventional diabetes therapy. The clinical findings further suggest that KWG may possess promising vascular benefits, offering a potential cost-effective natural health alternative. However, these results are preliminary and warrant long-term exploration.
I can’t believe how time flew by so quickly and here I am writing the acknowledgments section of my thesis. I joined the Nutritional Sciences graduate program in September 2012 as an international student having travelled all the way from Kuwait with the ultimate goal of earning a master’s degree. This experience, though challenging, has greatly enriched me on so many levels - academically, professionally, and personally. It has also offered me a great deal of meeting so many fantastic people, rendering my master’s studies experience pleasant, exciting, and memorable. It is for their collaborative significant contributions that this work was achievable. Thus, I am greatly indebted to these people and must acknowledge.

Firstly, I would like to express my sincerest gratitude and appreciation to my supervisor, mentor, and friend, Dr.Vuksan. Thank you for giving me the golden opportunity of carrying out a human clinical trial and trusting me in foreseeing this project. Your constant support, reassurance, and academic guidance along the way were vital in the completion of this work, and in shaping my interest towards clinical research. Having experienced every process of the human clinical trial right from scratch has broadened my vision in human research in ways I could have never imagined. Your constant kind sense of humor smoothened the so-many rough times and made the task much more bearable. I must add that your ongoing support, not only as a supervisor, but also as a close friend has given me the strength to overcome numerous obstacles along the road. I simply could not have asked for a more erudite, motivational, supportive, and kind-hearted academic supervisor. Thanks from the bottom of my heart!

Secondly, my deepest thanks goes to Dr.John Sievenpiper, whom I had the privilege of working with while carrying out my meta-analysis project. Your solid knowledge in the area, along with your supportive, kind character were inevitably crucial in the successful completion of the work. Thank you for being such a great inspiration, for enduring my never-ending questions,
and for teaching me the fundamentals of meta-analyses. You are truly a one-of-a-kind mentor and simply exceptional in every way.

I also extend my gratefulness to my advisory committee members, Dr. David Jenkins and Dr. Richard Bazinet, whom I was fortunate to be guided by throughout my master’s studies. Many thanks for offering such great guidance, for your constant encouragement and valuable feedback.

In addition, my wonderful friends, whom greatly contributed to the successful completion of this thesis, must be recognized. I’d like to begin by thanking the people from the risk factor modification center, the meta-analyses center: Adrian, Vanessa, Jay, Sonia, Effie, and Vivan, your constant assistance and beautiful spirit has made the experience worthwhile. Thank you simply for everything. As well, my colleagues and all the volunteer students that tremendously helped in the clinical trial phase deserve many thanks: Danielle, Shirley, Stacy, Allison, Thanh, Elena, Andreea, and Rodney, thank you all for being such great friends and showing much support. A special thanks also goes to all the study participants without whom this work would have never been possible.

Finally, and most importantly, I owe my master’s success to my amazing family. Dad, mom, Laila, Sara, Husain, and Hadi: I know that I wouldn’t have been able to be where I am today if it weren’t for each and every one of you. I am extremely grateful for your continuous support, encouragement, and unconditional love. Last but not least, my greatest gratitude goes to Kuwait University for sponsoring my master’s studies and allowing me to pursue my passion in research.
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LIST OF ABBREVIATIONS

AG- American Ginseng
AI- Augmentation index
AACE- American Association of Clinical Endocrinologists
AMPK- Adenosine monophosphate-activated protein kinase
ADA- American Diabetes Association
ABP- Ambulatory blood pressure
ABPM- Ambulatory blood pressure monitor
BMI- Body mass index
BP- Blood pressure
BBP- Brachial blood pressure
CAM- Complementary and alternative medicine
CVD- Cardiovascular disease
CDA- Canadian Diabetes Association
CBP- Central blood pressure
C- Compliance
cGMP- Cyclic guanosine monophosphate
D- Distensibility
DBP- Diastolic blood pressure
FPG- Fasting plasma glucose
FBG- Fasting blood glucose
FPI- Fasting plasma insulin
GDM- Gestational diabetes mellitus
GLUT 4- Glucose transporter isoform 4
HbA1c- Glycated hemoglobin
HPLC - High performance liquid chromatography
HOMA-IR - Homeostasis model assessment of insulin resistance
IGT - Impaired glucose tolerance
IFG - Impaired fasting glucose
iAUC - Incremental area under the glucose curve
KRG - Korean red ginseng
KWG - Korean white ginseng
MQS - Heyland methodological quality score
NHP - Natural health product
NEFA - Non esterified fatty acids
NGT - Normal glucose tolerance
OGTT - Oral glucose tolerance test
PPBG - Postprandial blood glucose
PWV - Pulse wave velocity
PPT - Protopanaxatriol
PPD - Protopanadiol
RCT - Randomized controlled trial
SBP - Systolic blood pressure
T2DM - Type 2 diabetes mellitus
T1DM - Type 1 diabetes mellitus
T1/2 - Half-life
WHO - World Health Organization
75g-OGTT - 75g Oral glucose tolerance test
2hPG - 2-hour plasma glucose
CHAPTER 1

INTRODUCTION
1. INTRODUCTION

The incidence of type 2 diabetes mellitus (T2DM) has been rising at an alarming rate over the last 2 decades and it is now considered to be a global epidemic. Latest statistics indicate that it affects 382 million people worldwide, with projections of a 55% increase by 2035\(^1\). T2DM compounds an individual’s risk for developing cardiovascular disease (CVD)\(^2\). Prolonged hyperglycemia and vascular abnormalities, the physiological hallmarks of diabetes, result in diabetes related micro- and macro-vascular complications\(^3\). As such, tight glycemic control and well managed blood pressure (BP) are fundamental to achieving optimal diabetes care\(^4\). However, these treatment goals generally go unmet\(^5\), as recent estimates revealed that only half of the diagnosed individuals with diabetes are meeting their glycemic targets, and only 36% are achieving goals for BP\(^6\). The progressive nature of the disease, along with the unfulfilled treatment challenges generates a compelling argument for safe, effective, and affordable alternative treatments that can be used as an adjunct to current medical therapies. Concurrent with the increasing demand for more effective medications is the recent upsurge in the use of herbal remedies amongst the general public\(^7\). Research estimates that there has been a 3-fold increase in the consumption of herbal medicine in the United States, and that more than one third of Canadians are using some form of complementary and alternative medicine (CAM)\(^8\).

Ginseng, a traditional medicinal plant, embodies an important position in the oriental pharmacopeia. Traditionally it is used primarily for treating illness, restoring homeostasis, and promoting longevity\(^9\), but more recently is has been identified as the most commonly used herbal for controlling CVD risk factors\(^{10}\). Of the thirteen ginseng species identified, two have been well documented in history and used as therapeutic agents: American ginseng (\textit{Panax quinquefolius}), and Asian ginseng (\textit{Panax ginseng})\(^{11}\). Data from multiple controlled trials using American or Korean ginseng have demonstrated its advantages for glycemic control, suggesting that it may be a promising therapeutic herb\(^{12-15}\). However, the consistency of clinical data is lacking, and there is overall insufficient evidence to support its anti-hyperglycemic efficacy. Four systematic reviews of
controlled clinical trials investigating the clinical efficacy of ginseng on blood glucose regulation amongst other health parameters revealed mixed findings: two concluded that there was a lack of convincing evidence for benefit\textsuperscript{16,17}, while two recent ones reported promising results for improving glucose metabolism\textsuperscript{18,19}. A systematic review and meta-analysis focusing solely on Korean red ginseng (steamed \textit{Panax} Ginseng variety) as a potential remedy for T2DM did not demonstrate favorable outcomes, which the authors attributed to the small number of studies with variable treatment regimens\textsuperscript{20}. Due to the inconsistency in the literature, along with the limited scope of data used to evaluate the role of ginseng in diabetes management, the evidence to support an anti-hyperglycemic indication for ginseng is limited. Accordingly, there is a clear need for more comprehensive and higher quality evidence to support the glycemic-lowering effects of ginseng for guidelines development.

Additionally, and more specifically, varieties of Korean red ginseng (KRG), a type of steamed Asian ginseng, have been shown to improve glycemic control and certain cardiovascular parameters when used in addition to conventional medication\textsuperscript{15,21,22}. However, despite being the most popular form of ginseng used in traditional dishes\textsuperscript{23}, scant clinical evidence exists on the health benefits of the white, non-steamed cultivars of Korean ginseng, Korean white ginseng (KWG). It has been suggested that steaming results in a more pronounced biological effect compared to raw dried ginseng\textsuperscript{24}, however the process significantly alters the profile of dammarane type saponins, a pharmacomologically active fraction of ginseng, also known as ginsenosides \textsuperscript{25}, and also results in the loss of malonyl type ginsenosides\textsuperscript{26,27}, all of which may possibly alter its therapeutic benefits. Given the favorable metabolic properties of KWG seen in animal models\textsuperscript{28,29}, along with the added cost of steaming of KRG\textsuperscript{11}, KWG may hold promising potential in improving certain CVD risks, and thus, serve as a less expensive therapeutic alternative to KRG. As clinical data on its medicinal properties is relatively scarce, there is a compelling need for exploring its CVD health benefits.
This thesis, therefore, aims at providing more comprehensive and higher quality evidence on ginseng’s anti-hyperglycemic potential through the conduction of a systematic-review and meta-analysis of RCTs of the effect of ginseng on glycemic control in individuals with and without diabetes. It also attempts to clinically assess the glycemic and vascular health effects of KWG via undertaking a dose response crossover trial in subjects with T2DM. Whilst a linear dose response effect is not expected to be seen, as observed previously in similar study designs with ginseng\textsuperscript{30}, a dose response design will aid in identifying a potential optimal therapeutic dose that can serve as pilot data for possible long term exploration.
CHAPTER 2

LITERATURE REVIEW
2. LITERATURE REVIEW

This review of the literature aims at discussing the subject of ginseng and its potential
glycemic and vascular health benefits in context of the escalating prevalence of T2DM and its
associated CVD risks.

2.1 The diabetes epidemic

2.1.1 Prevalence and global burden

Diabetes has become one of the major public health challenges and is approaching
epidemic proportions globally. It currently affects 382 million people worldwide, and is
predicted to rise to 592 million by 2035. In Canada, more than 2.7 million are living with
diabetes, and this number is expected to reach 4.2 million by 2020. Although the number of
Canadians diagnosed with diabetes is already high, an additional almost 1 million are estimated
to have undiagnosed diabetes. Of the total number of affected individuals with diabetes, 80% are
living in low- and middle-income countries. The human, social, and economic burden of
diabetes is huge, being responsible for 5.1 million deaths, and a global health expenditure of
$548 billion in 2013. With the rising number of cases, the decreased quality of life and life
expectancy, and the associated health care costs, much effort is being put into both its prevention
and treatment. However, despite myriad tools and strategies used to prevent and halt the
complications of this disease, the prevalence of diabetes still remains high and the major
therapeutic goals go unmet.

2.1.2 Definition of diabetes

Diabetes mellitus is a metabolic disorder illustrated by elevated blood glucose levels due
to defective insulin secretion, insulin action or both. Long term hyperglycemia is associated with
relatively specific long-term microvascular complications affecting the eyes, kidneys and nerves,
as well as an increased risk for CVD. The basis upon which diabetes is diagnosed are thresholds of glycemia that are related to microvascular disease, especially retinopathy\textsuperscript{34}.

2.1.3 Classification of diabetes

There are several etiologically based classifications of diabetes. These include type 1, type 2, gestational and other specific types of diabetes. Following will briefly describe each type.

2.1.3.1 Type 1 diabetes mellitus

Type 1 diabetes mellitus (T1DM), formerly known as insulin dependent diabetes, includes those type of diabetes that are prone to ketoacidosis and are associated with an absolute insulin deficiency as a result of pancreatic $\beta$-cell destruction through either an autoimmune process or a process for which an etiology is not known. It is thought that both environmental and genetic factors are involved in its development. Environmental factors may include early exposure to foreign proteins from cow’s milk based infant formulas or wheat. To date, there is no convincing evidence for a role of modifiable lifestyle risk factors in the development of type 1 diabetes\textsuperscript{35}.

2.1.3.2 Type 2 diabetes mellitus

T2DM, is a progressive metabolic disorder characterized by inadequate insulin production and insulin resistance\textsuperscript{36}, and is responsible for $\sim$90-95\% of the cases of diabetes globally\textsuperscript{35}. T2DM is thought to have some genetic predisposition, but is also largely determined by lifestyle factors. It is often seen in obese individuals with a sedentary lifestyle.

2.1.3.3 Gestational diabetes mellitus

Gestational diabetes mellitus (GDM) is characterized by elevated glucose levels with onset or first recognition during pregnancy, which generally returns to normal status after birth.
GDM complicates ∼4% of all pregnancies in the U.S., resulting in ∼135,000 cases annually. GDM represents nearly 90% of all pregnancies complicated by diabetes.\textsuperscript{35}

\subsection*{2.1.3.4 Other specific type of diabetes}

Other specific types include a wide variety of relatively uncommon conditions, primarily specific genetically defined forms of diabetes or diabetes associated with other diseases or drug use\textsuperscript{34}.

\subsection*{2.1.3.5 Intermediate stages of hyperglycemia: pre-diabetes}

Pre-diabetes is a practical term referring to impaired fasting glucose (IFG), impaired glucose tolerance (IGT) or a glycated hemoglobin (HbA1c) of 6.0% to 6.4%, each of which places individuals at high risk of developing T2DM and its complications. IFG and IGT represent intermediate states of abnormal glucose regulation that exist between normal glucose homeostasis and diabetes. The Canadian Diabetes Association (CDA) defines IFG as a fasting plasma glucose (FPG) value of 6.1 mmol/L (109.9 mg/dL) to 6.9 mmol/L (124.3 mg/dL) due to the higher risk of developing diabetes in these individuals compared to defining IGT as a FPG value of 5.6 mmol/L (100.9 mg/dL) to 6.9 mmol/L (124.3 mg/dL) \textsuperscript{34}. According to the American diabetes Association (ADA), IFG is defined as having FPG levels of 5.6 mmol/L (100.9 mg/dL) to 6.9 mmol/L (124.3 mg/dL), while IGT is characterized by having blood glucose levels of 7.8 mmol/L (140.5 mg/dL) to 11.0 mmol/L (198.2 mg/dL) after 2 hours of an oral glucose tolerance test (OGTT)\textsuperscript{35}.

\subsection*{2.1.4 Diagnostic criteria for diabetes}

There are 3 relevant sets of international guidelines for the diagnosis of diabetes: the World Health Organization (WHO), CDA, and ADA. New cases of T1DM, T2DM, pre-diabetes including IFG and IGT in non-pregnant individuals are diagnosed by FPG, random, or postprandial (2hr plasma glucose during a 75g oral glucose tolerance test [75g-OGTT]).
Apparent classic symptoms including polyuria, polydipsia, ketonuria, rapid weight loss, polyphagia, and blurred vision are also included in the diagnosis\textsuperscript{34}. Table 1 outlines the diagnostic criteria based on CDA and ADA.

**Table 1. Canadian Diabetes Association and American Diabetes Association diagnostic criteria for diabetes in non-pregnant adults**

<table>
<thead>
<tr>
<th>Test</th>
<th>Result</th>
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<tbody>
<tr>
<td>FPG</td>
<td>$\geq 7 \text{ mmol/L (121 mg/dL)}$</td>
</tr>
<tr>
<td>HbA1c</td>
<td>$\geq 6.5%$</td>
</tr>
<tr>
<td>2hPG in a 75g-OGTT</td>
<td>$\geq 11.1 \text{ mmol/L (200 mg/dL)}$</td>
</tr>
<tr>
<td>Random PG</td>
<td>$\geq 11.1 \text{ mmol/L (200 mg/dL)}$</td>
</tr>
</tbody>
</table>

In the absence of symptomatic hyperglycemia, if a single laboratory test result is in the diabetes range, a repeat confirmatory laboratory test must be done on another day. In the case of symptomatic hyperglycemia, a confirmatory test is not required. 2hPG, 2-hour plasma glucose; HbA1c, glycated hemoglobin; FPG, fasting plasma glucose; OGTT, oral glucose tolerance test; PG, plasma glucose. Derived from CDA and ADA diagnostic criteria.

### 2.1.5 Assessment of glyceemia in type 2 diabetes

#### 2.1.5.1 Postprandial blood glucose

Postprandial blood glucose (PPBG) relates to plasma glucose concentrations after food intake, and its profile is established by many factors. In those without diabetes, fasting plasma glucose concentrations (i.e., following an overnight 8 to 10-hr fast) generally range from 3.9 mmol/L (70 mg/dL) to 6.1 mmol/L (110 mg/dL)\textsuperscript{37}. Rise in glucose concentrations begin $\sim 10\text{min}$ after the beginning of food intake due to dietary carbohydrate absorption. Many factors determine the PPBG profile including carbohydrate absorption, insulin and glucagon secretion, and their coordinated effects on glucose metabolism in the liver and peripheral tissues\textsuperscript{38}. The degree and peak time of plasma glucose concentration depend on a variety of factors, including the timing, quantity, and composition of the meal.
In individuals without diabetes, peak in plasma glucose concentration is generally observed ~60 min after the start of a meal, seldom go above 7.8 mmol/L (140 mg/dL), and return to preprandial levels within 2–3 hrs. On the other hand, those with T2DM are characterized by impairment of the early-phase insulin release, which generally occurs early in the development of the disease\textsuperscript{39}. Accordingly, individuals with IGT experience a delay in peak insulin levels, while in those with T2DM, there is both a delay in peak insulin levels, along with inadequate insulin levels to manage PPBG excursions effectively\textsuperscript{40}. Generally, PPBG excursions are higher and more extended in people with T2DM compared to those without diabetes. This is due to abnormalities in insulin and glucagon secretion, hepatic glucose uptake, suppression of hepatic glucose production, and peripheral glucose uptake. As food absorption continues for 5–6 hrs after a meal in those both with or without diabetes, the optimal time to measure postprandial glucose concentration must be determined\textsuperscript{41}. Generally, a plasma glucose measurement at 2 hrs after the start of a meal is sensible, as it usually approximates the peak value in patients with diabetes, and provides a reasonable assessment of postprandial hyperglycemia. Specific clinical conditions, such as gestational diabetes or pregnancy complicated by diabetes, may benefit from testing at 1 hr after the meal\textsuperscript{41}.

As aforementioned, the early-phase insulin response to glucose is reduced in those with IGT, and is both delayed and blunted in those with T2DM\textsuperscript{40}. Consequently, the lack of early-phase insulin release throughout and following the prandial phase has numerous deleterious effects on normal glucose homeostasis such that hepatic glycogenolysis and gluconeogenesis are not inhibited sufficiently, and glucose uptake by muscle is insufficient. This as a result leads to postprandial hyperglycemia observed in individuals with glucose intolerance and T2DM\textsuperscript{42}. Recognizing these pathophysiologic changes is essential since they indicate that elevated postprandial glucose is different from elevated fasting glucose. It also shows that elevations in PPBG appear to be a sign of the earliest abnormalities that can be detected and potentially controlled/managed in order to stop or inhibit the process leading to \( \beta \)-cell failure.
2.1.5.2 Postprandial blood glucose as a marker of cardiovascular disease risk

Up until recently, the precise role of PPBG to the overall glycemic control of patients with T2DM remained largely unclear. Generally, both HbA1c and FPG have been regarded as valid markers for overall glucose exposure, and therefore were routinely used to evaluate glycemic control and management in T2DM. Yet, studies conducted more recently, have suggested that a third element of the glucose triad, the PPBG excursions, might have a role in the overall glycemic load and might also be a sign of glycemic management. Evidence for this stems from numerous epidemiological studies that have been conducted in the past 20 years. These investigations including, but not limited to, the Hoorn study, the Honolulu study, the Chicago Heart study, and the Diabetes Epidemiology: Collaborative analysis of Diagnostic Criteria in Europe (DECODE) study, have demonstrated a relation between the 2-hr plasma glucose, following a 75g OGTT, and the prevalence of CVD. A further analysis of the DECODE data focusing on CVD (268,811 person-years) showed that PPBG is an independent predictor of CVD mortality as opposed to FPG. This was also seen in another study which showed that postprandial glycemia, but not fasting glucose, predicts infarction in T2DM. Confirmation for this association was also provided by a meta-analysis of 20 studies involving 95,783 individuals who experienced 3,707 cardiovascular events over 12.4 years. Collectively, these data offer support for the notion that postprandial hyperglycemia in subjects with IGT or T2DM is a powerful marker of CVD, and that any persistent elevation in glycemia (dysglycemia) is a cardiovascular risk factor. Given that PPBG appears to be the earliest dysglycemic marker for CVD risk, it therefore seems very sensible to introduce it to the list of the recognized continuous CVD risk factors, such as BP and LDL-cholesterol, where treatments should aim at specifically lowering PPBG, as emphasized by the ADA.
2.1.5.3 Glycated hemoglobin as a marker of long term glycemic health

Presently, HbA1c measurements are the “gold standard” for evaluating long-term glycemic health. It is the extent to which hemoglobin is glycosylated in erythrocytes and is expressed as a percentage of total hemoglobin concentration. It reflects the exposure of erythrocytes to glucose in an irreversible time- and concentration-dependent manner. HbA1c levels provide a measure of the average blood glucose concentration during the preceding 2–3 months. Due to the high variation in blood glucose concentrations throughout a 24-hr period and from day to day in diabetes, the assessment of HbA1c is the most accepted marker of long-term glycemic control. Today, HbA1c is the most widely used marker for glycemic control, showing a close relation to both risks of macrovascular, and microvascular complications, in comparison to single or episodic glucose levels. In 2009, The International Expert Committee recommended the use of HbA1c to diagnose diabetes mellitus with a threshold >6.5%. Yet, despite having its merits, numerous factors have been found to affect HbA1c levels. People with certain medical illnesses, like kidney disease or infections will have abnormal red blood cell survival and will tend to have a lower HbA1c, interfering with the use of HbA1c in understanding their glucose status. Ethnic origin may also play a role as a number of studies have shown that African Americans have a higher level of HbA1c for a given level of blood sugar than do non-Hispanic whites. Additionally, age is another factor that affects HbA1c. Genetics has also been found to affect HbA1c as there are likely genetic differences from one person to another which cause one person to be a ‘high glycator’ and another person to be a ‘low glycator’.

2.1.6 Pathophysiology of Type 2 diabetes mellitus

The pathogenesis of T2DM can be characterized by a cluster of complications including peripheral insulin resistance, impaired regulation of hepatic glucose production, and a decline in β-cell function, which would ultimately lead to β-cell failure. Understanding the pathophysiology of T2DM requires a conceptualization of the framework within which glycemia is
controlled. Regulation of blood glucose is attained by the key hormone, insulin. Normoglycemia is achieved by the balanced interplay between insulin action and insulin secretion. Importantly, the normal pancreatic β-cell can adapt to changes in insulin action, where a decrease in insulin action is followed by upregulation of insulin secretion (and vice versa)\(^6\). The curvilinear relation between normal β-cell function and insulin sensitivity is illustrated in Figure 1. In patients with IGT and T2DM, deviation from this hyperbola results when β-cell function is insufficiently low for a certain level of insulin sensitivity. Thus, β-cell dysfunction is a critical element in the pathogenesis of type 2 diabetes\(^6\). Yet, glycemia is not only affected by deviation from, but also progression along the hyperbola. When a decline in insulin action is seen, as in those with obesity, the system usually compensates by increasing β-cell function. However, at the same time, concentrations of blood glucose at fasting and 2hr after glucose load will modestly increase. This rise may be insignificant, but over time becomes destructive because of glucose toxicity, and in itself a cause for β-cell dysfunction\(^6\). Thus, even with (theoretically) unlimited β-cell reserve, insulin resistance sets the basis for the occurrence of hyperglycemia and T2DM\(^6\).

**Figure 1. Hyperbolic relation between β-cell function and insulin sensitivity**

![Hyperbolic relation between β-cell function and insulin sensitivity](image)

IGT: Impaired glucose tolerance; NGT: Normal glucose tolerance; T2DM: Type 2 diabetes
2.1.6.1 Insulin resistance

Insulin resistance is said to be present when the biological action of insulin is not meeting the target goals of both disposing glucose in the skeletal muscle and suppressing endogenous production of glucose primarily in the liver\textsuperscript{70}. In the fasting state, however, muscle accounts for only a small proportion of glucose disposal (<20%) while endogenous glucose production accounts for all the glucose entering the plasma. Endogenous glucose production is increased in those with T2DM or IFG\textsuperscript{71,72}. Because this rise occurs in the existence of high levels of plasma insulin, at least in the early and intermediate disease phases, hepatic insulin resistance is the driving force of hyperglycemia of T2DM\textsuperscript{69} (Figure 2).

2.1.6.2 Obesity and insulin resistance

A strong and clear association exists between insulin resistance and obesity. Numerous mechanisms mediating this relation have been established. A number of circulating hormones, cytokines, and metabolic fuels, such as non-esterified fatty acids (NEFA) originate in the adipocyte and alter the function of insulin. Elevated levels of stored triglyceride, especially in visceral or deep subcutaneous adipose depots, result in large adipocytes that are, by their own, resistant to the ability of insulin to suppress the breakdown of lipids (lipolysis). This leads to increased release of circulating levels of NEFA and glycerol, both of which exacerbate insulin resistance in skeletal muscle and liver\textsuperscript{73} (Figure 2). Increased the storage of fat in non-adipose cells also plays a significant role\textsuperscript{74}. For example, under specific conditions, increased intramyocellular lipids are associated with skeletal muscle insulin resistance\textsuperscript{75}. The relation between intra-hepatic lipids and hepatic insulin resistance appears to be even closer\textsuperscript{76}. 
2.1.6.3 β-Cell dysfunction

Despite the fact that insulin resistance is considered the starting point in the pathogenesis of T2DM, pancreatic β-cell dysfunction is fundamental in the development of the disease and hyperglycemia does not become apparent until there is extreme β-cell damage. Throughout the initial stages in the normal history of T2DM during which negligible variations in FPG can be seen in the face of insulin resistance, elevated insulin levels respond as a mean of compensatory mechanism. As insulin resistance deteriorates with progression from normal glucose tolerance (NGT) to an IFG/IGT state, β-cell function is further diminished, and eventually results in overt diabetes. β-cell function decline is specifically obvious when insulin secretion is expressed in relation to ambient insulin sensitivity. Subjects with IFG/IGT already have a 50% to 75% decline in β-cell function, in comparison to those with healthy NGT. Added progressive decline in β-cell function is seen over the subsequent years following the diagnosis of diabetes.
2.1.7 Type 2 diabetes and cardiovascular disease

Diabetes has become one of the chief risk factors for CVD. Vascular complications associated with diabetes include retinopathy and nephropathy, peripheral vascular disease, stroke, and coronary artery disease\(^{81}\). In addition, diabetes has detrimental effects on the heart muscle, resulting in both systolic and diastolic vascular complications\(^{81}\). The etiology of this excess cardiovascular morbidity and mortality is not completely clear. Data in the literature shows that though hyperglycemia, the hallmark of diabetes, results in myocardial damage following ischemic events; it does not represent the sole factor. This is because both pre-diabetes and the metabolic syndrome, even in patients with normal glycemia, augment the risk of most types of vascular diseases\(^{57,82,83}\). Decreasing the risk of CVD in subjects with T2DM, by means of improving glycemic control, managing dyslipidemia, and controlling hypertension, is the focus of health care systems.

2.1.7.1 Arterial stiffness as a strong predictor of cardiovascular disease risk

Arterial stiffness has become increasingly recognized as a powerful predictor of CVD\(^{84,85}\). It illustrates the diminished ability of an artery to expand and contract in response to changes in pressure. Arterial stiffness increases progressively with age in populations around the world\(^{86}\). Important indices that indicate the stiffening of the vessels include compliance and distensibility. Compliance (C) is a measure of volume change (∆V) in response to a change in blood pressure (∆P; C = ∆V/∆P). When a vessel is stiffened, the compliance as a result of volume change, is reduced for any given pressure change. However, compliance is also linked to initial arterial volume because a smaller volume reduces compliance for any given elasticity of the arterial wall. Distensibility (D) is compliance relative to initial volume (D = ∆V/∆P × V), making it more closely related to wall stiffness\(^{87}\). Increased propagation velocity of the pressure pulse along the arterial tree, called pulse wave velocity (PWV), results as a consequence of reduced compliance/distensibility. A number of different methods for the assessment of arterial stiffness
have been identified, some of which are more widely applicable in the clinical setting than others. This includes pulse pressure, PWV, ultrasound-derived indices, MRI-derived indices, and waveform analysis\(^9^8\). As PWV is now widely accepted as the ‘gold standard’ measure of arterial stiffness, it will be discussed in the following section.

2.1.7.1.1 Pulse wave velocity as a measure of arterial stiffness

The assessment of pulse wave velocity is generally accepted as the most simple, non-invasive, robust, and reproducible method to measure arterial stiffness\(^8^9\). It represents the velocity at which the forward pressure wave is travelled from the aorta through the vascular tree. Its calculation involves measuring the time taken for the arterial waveform to pass amongst two points a measured distance apart, and requires recording the readings from the two sites simultaneously, or gating separate recordings to a fixed point in the cardiac cycle, usually the R wave of the electrocardiography\(^9^0\). The Bramwell and Hill equation, which relates PWV to distensibility, can be described as the increased propagation velocity of the pressure pulse along the arterial tree as a consequence of reduced compliance/distensibility. The equation is as follows: \(PWV = \sqrt{V \times \frac{\Delta P}{\rho \times \Delta V}}\), where \(\rho\) is blood density, and \(\Delta PV/\Delta V\) is relative volume elasticity of vessel segment.\(^9^1\). Various different methods have been used, both invasive and non-invasive, and can be applied to either flow or pressure waves. In light of the predictive power of PWV in determining CVD risk\(^9^2\), strategies that may potentially prevent or reduce stiffening, and thus, lower the risk of CVD events, are becoming the focus of attention.

2.1.7.1.2 Augmentation index as a surrogate marker of arterial stiffness

Wave reflection from the periphery to the central blood vessels augments the aortic pressure wave. An incident wave travelling from the left ventricle to the periphery reaches smaller vessels which act as a mirror, reflecting it back to the aorta. Thus, the resulting pressure in the ascending aorta is the sum of the incident and reflected wave. The effect of the reflected wave
on the incident wave in the ascending aorta is expressed as the augmentation index (AI), and is a measure of the additional load that the left ventricle is subject to due to wave reflection. AI is calculated as the height of the reflected wave as a percentage of the total height of the ascending aortic pressure wave. AI, derived from analysis of the radial pulse wave, is a simple and convenient method to measure arterial stiffness, and therefore has been widely used in clinical studies. AI is dependent upon the length of the cardiac cycle (which depends on the heart rate), the duration of systole and diastole, the speed of the pulse wave, the incident waveform, and the amplitude of the reflected pulse wave. For a given heart rate, the time of arrival of the reflected wave depends on the speed of PWV, which is determined by the stiffness of the vasculature (PWV increases with increasing vessel stiffness). If the reflected wave arrives early in the cardiac cycle it combines with the incident wave, giving a greater ascending aortic pressure against which the left ventricle has to pump. However, if it arrives later in the cardiac cycle it increases the ascending aortic pressure in diastole leading to improved coronary circulation. If the cardiac cycle is longer (i.e., the heart rate is slower) the reflected wave is more likely to arrive in systole. As AI is dependent on PWV, which in turn is dependent on arterial stiffness, it is no surprise that AI has been proposed as a marker for arterial stiffness.

2.1.8 Management of type 2 diabetes mellitus

Successful management of T2DM requires strict control of glycemia as well as other risk factors to prevent disease progression. Consequently, preventing long term complications of T2DM must be the basis upon which therapeutic options are made. Due to the fundamental role of insulin resistance in the pathogenesis of T2DM and especially its adverse cardiovascular outcomes, interventions should originally be aimed towards improvement in tissue insulin sensitivity. This includes lifestyle modification, along with exercise and weight loss, which has shown an obvious decline in the progression of IGT to overt diabetes. It has also demonstrated improvements in numerous cardiovascular risk markers associated with the metabolic syndrome. A new
comprehensive T2DM treatment algorithm, being the first to include obesity, prediabetes, and CVD risk management, was issued by the American Association of Clinical Endocrinologists (AACE)\textsuperscript{100}. Due to the clear link between weight loss and improvements in blood glucose status, obesity management was incorporated in the algorithm\textsuperscript{98}. The level of treatment depends on the original HbA1c levels, as in AACE’s previous glycemic-control algorithm. Lifestyle modification, including weight loss, is a component of all treatments. The current algorithm recommends an HbA1c of 6.5% or less for healthy patients without concurrent illness and who are at low risk but individualized target HbA1c values greater than 6.5% for patients with concurrent illness and those who are at risk for hypoglycemia. This is unlike the earlier algorithm where an HbA1c of 6.5% or lower was recommended as the goal for most patients.

2.1.9 Pharmacotherapy used in type 2 diabetes mellitus and an opportunity for complementary and alternative medicine

Current pharmacological treatments available for T2DM consist of sulfonylureas and related agents, including biguanides, thiazolidinediones, α-glucosidase inhibitors and insulin. Of the most common side effect of sulfonylurea is hypoglycemia, which though usually not very severe, could cause fatal complications\textsuperscript{101,102}. The main adverse effects with thiazolidinediones is weight gain (depending on dose) of 1-4 kg following 6 months of use\textsuperscript{103}. In hopes of overcoming the side-effects that are associated with oral hypoglycemic drugs, a new class of drugs called gliptins or dipeptidyl peptidase 4 (DPP-4) inhibitors was introduced. They work by extending the action of gut hormones called incretins, which enhance insulin levels. The biggest advantage of the gliptins seems to be their potential to stimulate insulin production with minute chances of hypoglycemic episodes. However, despite the availability of multiple classes of oral antidiabetic drugs and insulin, as well as the new pharmacological approaches to improve glycemic control, the majority of patients fail to attain or maintain tight glycemic management over time, raising their risk of serious microvascular and macrovascular complications\textsuperscript{104}. This as a result has
increased people’s interest towards seeking other strategies to better control their glycemic status devoid of the associated side effects. Of these is the use of complementary and alternative medicine (CAM)\textsuperscript{105}, which will be discussed herein.

2.1.10 Complementary and alternative medicine in the management of type 2 diabetes mellitus

Based on the National Center for Complementary and Alternative Medicine, USA, CAM is a group of diverse medical and healthcare systems, practices, and products that are not presently considered to be part of orthodox medicine. CAM can be classified into 5 major domains: alternative medical systems, mind-body interventions, biologically-based therapies, manipulative and body-based methods, and energy therapies\textsuperscript{106}. Biologically-based therapies are defined as therapies using substances found in nature, such as dietary supplements, and herbal and botanical products, and they have been reported to be the most frequently used form of CAM among individuals with chronic illnesses including diabetes and CVD\textsuperscript{105,107}. More specifically, treatment for T2DM is an active area of research for alternative medicines, to supplement or possibly replace current drug treatments. Of particular interest has been the use of traditional herbal medicines. In recent years ginseng, a root of the genus \textit{Panax} \textsuperscript{108}, has gained much attention for this reason. Studies suggests that certain types of ginseng may have anti-hyperglycemic effects and can therefore be used in conjunction with traditional T2DM medication or therapy\textsuperscript{108}, although the type and preparation of the ginseng may vary its ability to lower postprandial glycemia due to variations in its ginsenoside profile\textsuperscript{109,110}. The following review will discuss ginseng and its major active components, with a focus on its role as a promising medicinal herb in the management of T2DM and CVD.
2.2 Ginseng as a potential therapeutic herb

2.2.1 Background

Ginseng, the root of the genus Panax, has a prolonged history of use in the far East, specifically in Korea and China, as a valued herbal medicine in maintaining physical vitality and restoring homeostasis. The genus name “Panax” was given by the Russian botanist, C.A. Meyer, and is derived from the Greek word (Pan=all + axos=medicine) meaning ‘cure all’. As well, the name ginseng comes from the Chinese term renshen, meaning man-root. It is thought to represent the three essences of man including body, mind, and spirit, and is recognized as the ‘king of herbs'. Ginseng has been mainly used as a tonic to revitalize weaknesses, but only recently as a therapeutic agent, even though, based on the ‘Encyclopedia of Herbs’ written by Li Shizhen in China, 1596 A.D., it was incorporated as a component for treating 23 diseases. It was also included in the Korean Clinical Pharmacopeia’, written by the Korean herbalist, Huh Joon in 1610 A.D., as a component present in 653 prescriptions.

2.2.2 Classification, description and processing

The ginseng plant is a deciduous perennial derived from several species that belong to the family Araliaceae and genus Panax, indigenous both to Asia and North America. Thirteen distinct species of ginseng have been identified with numerous different cultivars. Of those, the most popular include: Panax ginseng C.A Meyer, cultivated in Korea; Panax quinquefolius L (American ginseng [AG]), found in Southern Canada and the United States of America; Panax japonicas C.A. Meyer (Japanese ginseng), grown in Japan; Panax notoginseng (Sanchi ginseng) grown in China’s Yunnan province; and Panax vietnamensis, grown in vietnam. Panax ginseng is not a generic name, when used; it includes all ginsengs originated from Asian countries, i.e., Panax ginseng C.A. Meyer, Panax japonicus, Panax notoginseng (Sanchi ginseng), and Panax vietnamensis. The ginseng
plant can be described as a shade-loving, deciduous perennial having five-fingered leaves, small white flowers, red berries, and a yellowish-brown root. The part of the plant that is used for medicinal purposes is mainly the root, although active constituents are present in other fractions of the plant\textsuperscript{113}. There are two different ways of processing ginseng after harvest: air drying and steaming the roots; the former results in white ginseng and the latter produces red ginseng. Interestingly, after these two different processes, the roots differ in their content of saponins\textsuperscript{114}, and this may be the reason for the variable effects of different ginseng products. The so-called white ginseng is peeled and then air/sun dried to reduce the water content to 12\% or less. Drying in the sun bleaches the root to a yellowish-white color. Red ginseng on the other hand is ginseng that is not peeled but steamed and then air/sundried. Steaming gives the roots a glossy reddish-brown coloring\textsuperscript{11}. In the course of the steaming process, ginseng starch is gelatinized, causing a change in ginsenoside content\textsuperscript{24}. Particularly, Korean ginseng (\textit{Panax ginseng} C.A. Meyer) is classified into three types, depending on how it is processed: fresh ginseng (less than 4 years old), white ginseng (4–6 years old and dried after peeling), and red ginseng (harvested when 6 years old, steamed and dried)\textsuperscript{115}. The content of ginsenoside compounds differs between the red and white forms. Growing time also impacts ginsenoside content, with roots from plants older than five years being more potent than roots from one- to two-year-old plants.

\subsection*{2.2.3 Ginseng phytochemistry}

Ginseng contains many phytochemicals, of which three are considered to be its main components. These include the triterpene saponins (ginsenosides), ginsenans (polysaccharides), and panaxans (peptides). Each have been isolated and characterized from 13 ginseng species using various techniques\textsuperscript{116}, such as high performance liquid chromatography (HPLC)/mass spectrophotometry and HPLC/ultraviolet. As well, their pharmacological activities have been thoroughly investigated and compared to one another, with the triterpene saponins, commonly known as ginsenosides, showing the greatest activity\textsuperscript{9}.
Ginsenosides with a few exceptions share a similar basic structure, consisting of a saturated 1,2-cyclopentanoperhydrophenanthrene (sterane or gonane) steroid nucleus. Based on their skeleton of aglycones, they are classified into two groups, namely dammarane-type and oleanane-type. Further expanding on this, ginsenosides within the dammarane-type consist mainly of 3 types classified base upon their genuine aglycone moieties: protopanaxadiol (PPD), protopanaxatriol (PPT), and ocutillol. On the other hand, ginsenosides of the oleanane-type are classified according to their aglycone oleanolic acid\(^{117}\) (Figure 3). More specifically, the nomenclature of the dammarane-type ginsenosides, which consist of a dammarane skeleton (17 carbons in a four-ring structure) with a variety of sugar moieties (e.g. glucose, rhamnose, xylose and arabinose) attached to the C-3, C-6, and C-20 positions\(^{118}\), is presented as 'Rx', where the 'R' represents the root and the 'x' describes the chromatographic polarity in an alphabetical order\(^{119}\).

More than 30 ginsenosides have been recognized and classified into two categories: (1) the 20(S)-PPD including Rb\(_1\), Rb\(_2\), Rb\(_3\), Rc, Rd, Rg\(_3\), and Rh\(_2\), and (2) the 20(S)-PPT including Re, Rf, Rg\(_1\), Rg\(_2\), and Rh\(_1\). The distinction among the PPTs and PPDs is the carboxyl group that is present at the C-6 position in PPTs\(^{118}\). With regards to the quality and composition of the ginsenosides in the ginseng plant, it has been shown to be affected by a range of factors including the species, age, part of the plant, cultivation method, harvesting season and preservation method\(^{120}\). The overall ginsenoside content in ginseng is positively correlated to its age, reaching maximum levels at around 6 years\(^{120,121}\).
Figure 3. Chemical structure of the dammarane and oleanane-type ginsenosides

2.2.4 Interspecies differences in ginsenosides

Numerous differences in the profile of the ginsenosides exist amongst different ginseng species, many of which are used in the authentication of ginseng. Reports have shown that Sanchi and AG have higher total ginsenosides compared to Asian ginseng, while Siberian is free on any ginsenosides. Additionally, some ginseng species are devoid of certain ginsenosides. For example, AG does not contain ginsenosides Rf and Rg. The ootillol-type dammarane saponins are not present in Asian ginseng, and red Asian ginseng does not contain malonyl ginsenosides. Finally, the ratio of various ginsenoside classes and common individual ginsenosides also differ among species. Relative to Asian ginseng, AG is distinct in that it contains a ratio <1 of the ginsenoside Rg1/Re and Rb2/Rc. Differences in the ratio of the
ginsenosides Rb₁ and Rg₁, as well as the neutral and malonyl ginsenosides are also present. In both Asian and Sanchi ginseng the ratio of Rb₁:Rg₁ is usually from 1-3, while in AG it is 3-10. Asian ginseng also contains a smaller proportion of malonyl ginsenosides (~10% of total) compared to AG (~40% of total).

2.2.5 Ginseng pharmacokinetic

Both animal and human studies have explored the pharmacokinetics of different ginseng saponin compounds, or ginsenosides. It has been shown that in order to obtain observable levels of ginsenosides in the plasma, the ingested test compound doses are often required to be at the upper end of the pharmacological dose range. Yet, due to the high diversity and heterogeneity amongst the chemical structures of different ginsenosides, the pharmacokinetic profile of ginseng has not been completely understood. Studies also reveal that the ginsenoside absorption rate is low after oral administration, and doses of test compounds must be high in order to detect levels in plasma. Many factors have been shown to limit the intestinal absorption of ginsenosides. This includes: extensive metabolism in the gastrointestinal tract, poor membrane permeability, and low solubility of deglycosylated products. Data in the literature demonstrates that the bioavailability of the PPD group of saponins (ginsenosides Ra₃, Rb₁, Rd, Rg₃, and Rh₂) and of the PPT group of saponins (ginsenosides Rg₁, Re, Rh₁, and R₁) was found to be less than 5%. It also shows that the PPT saponins have better bioavailability than PPD saponins, which could possibly be explained by the faster degradation rate of the PPD saponins compared to the PPT saponins. Bioavailability of ginsenosides has been shown to be improved through the ingestion of high oral doses, and by changing the pharmaceutical formulation. Research also indicates that ginsenosides are rapidly absorbed and readily distributed in the tissues, as it was shown that time required for ginsenosides to reach maximum concentration in the plasma was less than 2-hrs. In rabbits, the
half-life ($T_{1/2}$) elimination of Rg1, Re and Rb2 were between 0.8 hr and 7.4 hr\textsuperscript{139} while in humans, the $T_{1/2}$ of the researched ginsenosides was generally less than 24 hr\textsuperscript{128,140}

### 2.2.6 Pharmacological effects of ginseng

An extensive number of clinical, laboratory and animal studies have been performed to investigate the actions and chemistry of ginseng. Much of this research has been performed in China, Korea, and Japan. The best-documented pharmacological effects of ginseng in humans have been demonstrated in the central nervous, cardiovascular, endocrine and immune systems\textsuperscript{141}. In addition, ginseng and its accountable components have demonstrated tremendous qualities that assist in the treatment and prevention of the metabolic syndrome, including blood glucose regulation, insulin secretion stimulation, and anti-obesity effects\textsuperscript{142}. They have also been ascribed antineoplastic, antistress, and antioxidant activity\textsuperscript{9}. The following section will provide pre-clinical and clinical data relating to ginseng's glycemic and vascular health benefits which supplied substantial evidence and basis for the intended clinical study, as detailed in chapter 5. Presently, there is limited clinical evidence on the non-steamed *Panax ginseng* C.A. Meyer.

#### 2.2.6.1 The effect of ginseng on glycemic control

##### 2.2.6.1.1 Pre-clinical Evidence

Mounting evidence from rigorously conducted cell and animal studies indicate that ginseng, and more specifically, its ginsenosides, possess anti-diabetic potential. Both PPD-type saponins (Rb\textsubscript{1}, Rb\textsubscript{2}, Rc, Rg\textsubscript{3}, Rh\textsubscript{2}, compound K, and PPT-type saponins (Re, Rg\textsubscript{1}, Rg\textsubscript{2}, and PPT) were shown to exhibit anti-diabetic activity in cell culture and animal studies. Some of the most promising data are illustrated in Table2 and Table3.

As shown in these tables, various ginseng roots, along with different doses of ginsenosides and their metabolites, such as compound K, administered to cell lines and animal models demonstrated a significant lowering effect in fasting and PPBG. Data from these studies suggest
four plausible mechanisms of action through which blood glucose is improved. These include: 1) modulation of insulin production and secretion 2) modulation of glucose metabolism, 3) modulation of glucose uptake, and 4) modulation of inflammatory pathway. Data supporting the role of ginseng or its active components in insulin production enhancement are numerous. It has been shown that AG and KRG stimulate insulin secretion in HITT15 cells and isolated rat pancreatic islets, respectively\textsuperscript{143,144}. Other studies also demonstrate that AG and KRG increase insulin production and secretion through inhibition of cytokine-induced β-cell apoptosis\textsuperscript{143,145}. A study by Lee et al. revealed that decreases in plasma glucose levels paralleled increases in plasma insulin levels, when Wistar rats were intravenously injected with ginsenoside Rh\textsubscript{2}\textsuperscript{146}. This effect was shown to be mediated by stimulating muscarinic M\textsubscript{3} receptors in pancreatic cells. The most potent insulin secretion stimulating action was shown by compound K, an active metabolite of PPD ginsenosides. Studies using HITT15 cells and primary cultured islets, showed that compound K ameliorated insulin secretion and this effect was entirely eliminated in the presence of diazoxide (K\textsuperscript{+} channel opener) or nifedipine (Ca\textsuperscript{2+} channel blocker). The insulin secretion stimulating activity of compound K was also established with an OGTT in ICR and db/db mice. It was concluded from these studies that compound K decreased plasma glucose levels by stimulating insulin secretion and this action was plausibly associated with an ATP-sensitive K\textsuperscript{+} channel\textsuperscript{147}. Several other mechanisms by which ginseng can improve glycemia were revealed. Chung et al. showed that ginseng radix can ameliorate hyperglycemia possibly by means of blocking the intestinal glucose absorption and inhibiting hepatic glucose-6-phosphatase, while ginseng rootlet can improve hyperglycemia through the up-regulation of adipocytic peroxisome proliferator-activated receptor-\gamma (PPAR-\gamma) expression as well as inhibiting intestinal glucose absorption in KKAy mice\textsuperscript{28}. As well, modulation of glucose metabolism by means of inhibiting hepatic glucose production were also reported by certain ginsenosides. These include ginsenosides Rb\textsubscript{2}, Re and Rg\textsubscript{1}, where they were shown to suppress the hepatic gluconeogenesis in H4IIE and HepG2 cells, respectively, through the activation of AMP-activated protein kinase (AMPK)\textsuperscript{148-150}. Additionally, administration
of either *P. notoginseng*<sup>151</sup>, Rg<sub>3</sub><sup>152</sup>, Re<sup>150</sup>, or 20(S)-PPT<sup>153</sup> in skeletal muscle cells or adipocytes demonstrated enhancement of glucose uptake through glucose transporter 4 (GLUT4) overexpression. A recent long-term study by Lee et al. demonstrated that KRG taken at the dose of 200 mg/kg/d for a 40-week duration enhanced insulin sensitivity in Otsuka Long-Evans Tokushima fatty rats by increasing expression of peroxisome proliferator-activated receptor-γ coactivator-1α, nuclear respiratory factor-1, cytochrome c, cytochrome c oxidase-4, and GLUT4<sup>154</sup>.

**Table 2. In vitro studies examining the effects of ginseng extract, individual ginsenosides and their metabolites on glycemic control in cell lines used in diabetes research.**

<table>
<thead>
<tr>
<th>Study</th>
<th>Ginsenosides</th>
<th>Dose</th>
<th>Cell type</th>
<th>Outcome</th>
<th>Suggested mechanism</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chang et al. 2007&lt;sup&gt;155&lt;/sup&gt;</td>
<td>Rg1</td>
<td>0.01-0.1 μM</td>
<td>Caco-2</td>
<td>↓ Glucose absorption across intestinal epithelium</td>
<td>Decreased expression of sodium/glucose cotransporter</td>
</tr>
<tr>
<td>Han et al. 2007&lt;sup&gt;156&lt;/sup&gt;</td>
<td>Compound K (PPD)</td>
<td>8 μM</td>
<td>HIT T15, Pancreatic Islets from male SD rats</td>
<td>↑ Insulin secretion</td>
<td>Inhibition of ATP sensitive potassium channels</td>
</tr>
<tr>
<td>Park et al. 2008&lt;sup&gt;157&lt;/sup&gt;</td>
<td>Rg3</td>
<td>5 μM</td>
<td>HIT T15</td>
<td>↑ Glucose-stimulated insulin secretion</td>
<td>Activates AMPK pathway which stimulates translocation of glucose transporter</td>
</tr>
<tr>
<td>Zhang et al. 2008&lt;sup&gt;158&lt;/sup&gt;</td>
<td>Re</td>
<td>10 μM</td>
<td>3T3 L1</td>
<td>↑ Glucose uptake</td>
<td>Promotes GLUT4 translocation by activating IRS1 independent of IR activation, inhibits JNK and NFkB, activates PI3K</td>
</tr>
<tr>
<td>Shang et al. 2008&lt;sup&gt;159&lt;/sup&gt;</td>
<td>Rb1</td>
<td>0.01-10 μM</td>
<td>3T3 L1</td>
<td>↑ Insulin mediated glucose uptake</td>
<td>Promote translocation of GLUT1 and GLUT4 by activating PI3K</td>
</tr>
<tr>
<td>Park et al. 2008&lt;sup&gt;160&lt;/sup&gt;</td>
<td>Rg1, Rb1</td>
<td>20 μM</td>
<td>MIN 6</td>
<td>↑ Glucose-stimulated insulin secretion</td>
<td>PKA activation which lead to increased mRNA and protein expression of IRS2</td>
</tr>
<tr>
<td>Kim et al. 2008&lt;sup&gt;143&lt;/sup&gt;</td>
<td>KRG extract</td>
<td>0.1-1.0 mg/mL</td>
<td>Pancreatic Islets from male Sprague-Dawley rats</td>
<td>↑ Insulin secretion</td>
<td>Inhibition of ATP sensitive potassium channels</td>
</tr>
<tr>
<td>Study</td>
<td>Ginsenoside</td>
<td>Dose</td>
<td>Animal model</td>
<td>Outcome</td>
<td>Suggested mechanism</td>
</tr>
<tr>
<td>-----------------------------</td>
<td>------------------------------</td>
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<td>------------------</td>
<td>----------------------------------------------</td>
<td>----------------------------------------------------------</td>
</tr>
<tr>
<td>Park et al. 2008(^{161})</td>
<td>Ginseng radix extract</td>
<td>50 µg/mL</td>
<td>Pancreatic Islets from male rats</td>
<td>↑ glucose-stimulated insulin secretion</td>
<td>↑ mRNA expression of glucokinase (acts as a glucose sensor)</td>
</tr>
<tr>
<td>Lin et al. 2008(^{162})</td>
<td>AG berry extract, Re</td>
<td>0.1-1.0 mg/mL</td>
<td>MIN 6</td>
<td>↑ Insulin secretion</td>
<td>↓ Oxidative stress which can lead to insulin resistance</td>
</tr>
<tr>
<td>Kim et al. 2010(^{152})</td>
<td>Rg3</td>
<td>0.1-5.0 µM</td>
<td>MIN 6N8</td>
<td>↓ β-cell apoptosis</td>
<td>Prevented palmitate-induced apoptosis</td>
</tr>
<tr>
<td>Lee et al. 2010(^{163})</td>
<td>Rc</td>
<td>100 and 200 µM</td>
<td>C2C12</td>
<td>↑ Glucose uptake</td>
<td>Activation of an insulin-independent AMPK signaling pathway</td>
</tr>
<tr>
<td>Lee et al. 2011(^{149})</td>
<td>Rb2</td>
<td>10 mM</td>
<td>H4IE</td>
<td>Palmitate-induced gluconeogenesis</td>
<td>Activation of an AMPK-dependent signal Pathway</td>
</tr>
<tr>
<td>Lee et al. 2011(^{164})</td>
<td>Rg3 and Re</td>
<td>1–10 µM</td>
<td>3T3-L1</td>
<td>↑ Glucose uptake and GLUT4</td>
<td>Activation of PI3K pathways involving an ↑ in IRS-1</td>
</tr>
<tr>
<td>Quan et al. 2012(^{150})</td>
<td>Re</td>
<td>20 µM</td>
<td>HepG2</td>
<td>Inhibits hepatic glucose production</td>
<td>Activation of AMPK pathway</td>
</tr>
</tbody>
</table>

Rg1, Rg3, Re, Rb1- conventional nomenclature of ginsenosides; Compound K- protopanaxadiol metabolite; PPD- protopanaxadiols; KRG- Korean Red Ginseng; AG- American Ginseng; ATP- adenosine triphosphate; AMPK- adenosine monophosphate-activated protein kinase; GLUT4- glucose transporter isoform 4; IRS1- insulin receptor substrate protein 1; IR- insulin receptor; JNK- c-Jun N-terminal kinase; NFκB- nuclear factor kappa B; PI3K- phosphatidylinositol-3-kinase; GLUT1- glucose transporter isoform 1; PKA- protein kinase A; IRS2- insulin receptor substrate protein 2

Table 3. In vivo studies conducted with diabetes rodent models examining the effects of administration of ginseng extract, ginsenosides and their metabolites

<table>
<thead>
<tr>
<th>Study</th>
<th>Ginsenoside</th>
<th>Dose</th>
<th>Animal model</th>
<th>Outcome</th>
<th>Suggested mechanism</th>
</tr>
</thead>
<tbody>
<tr>
<td>Attele et al. 2002(^{165})</td>
<td>Berry extract, Re</td>
<td>150 mg/kg i.p</td>
<td>ob/ob mice</td>
<td>↓ Fasting and postprandial glycemia</td>
<td>↑ Insulin-stimulated glucose disposal</td>
</tr>
<tr>
<td>Xie et al. 2005(^{166})</td>
<td>Re</td>
<td>20 mg/kg i.p</td>
<td>ob/ob mice</td>
<td>↓ Fasting and postprandial glycemia</td>
<td>↑ Insulin sensitivity</td>
</tr>
<tr>
<td>Liu et al. 2005(^{167})</td>
<td>Panax Ginseng root</td>
<td>125 mg/kg oral</td>
<td>Male Wistar rats</td>
<td>↓ Plasma glucose AUC and glucose insulin index</td>
<td>↑ Insulin sensitivity</td>
</tr>
<tr>
<td>Lee et al. 2006(^{146})</td>
<td>Rh2</td>
<td>1.0 mg/kg i.v.</td>
<td>Male Wistar rats</td>
<td>↓ Fasting glycemia</td>
<td>↑ Insulin secretion</td>
</tr>
<tr>
<td>Han et al. 2007(^{156})</td>
<td>Compound K (PPD)</td>
<td>10/20 mg/kg oral</td>
<td>ICR and db/db mice</td>
<td>↓ Fasting and postprandial glycemia</td>
<td>↑ Insulin sensitivity</td>
</tr>
<tr>
<td>Study</td>
<td>Compound/Extract</td>
<td>Dose/Species/Method</td>
<td>Effect</td>
<td>Comments</td>
<td></td>
</tr>
<tr>
<td>------------------</td>
<td>----------------------------------</td>
<td>---------------------</td>
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<td></td>
</tr>
<tr>
<td>Yoon et al. 2007</td>
<td>Compound K</td>
<td>10 mg/kg db/db mice</td>
<td>↓ Fasting glyceremia</td>
<td>↑ Insulin secretion</td>
<td></td>
</tr>
<tr>
<td>Lee et al. 2007</td>
<td>Rh2</td>
<td>1.0 mg/kg i.v. Male Wistar rats</td>
<td>↓ Fasting glyceremia</td>
<td>↑ Insulin sensitivity</td>
<td></td>
</tr>
<tr>
<td>Banz et al. 2007</td>
<td>Root extract</td>
<td>10 g/kg Male ZDF rats</td>
<td>↓ Postprandial glyceremia</td>
<td>↑ Insulin sensitivity</td>
<td></td>
</tr>
<tr>
<td>Xie et al. 2007</td>
<td>AG berry juice</td>
<td>0.6 mL/kg ob/ob mice</td>
<td>↓ Postprandial glyceremia</td>
<td>Not specific</td>
<td></td>
</tr>
<tr>
<td>Chen et al. 2008</td>
<td>Rg1, Rb1, Rd, Re</td>
<td>50/200 mg/kg KK-Ay male mice</td>
<td>↓ Fasting and postprandial glyceremia</td>
<td>↑ Insulin sensitivity</td>
<td></td>
</tr>
<tr>
<td>Park et al. 2008</td>
<td>Rg3</td>
<td>12.5,25 mg/kg oral ICR mice</td>
<td>↓ Postprandial glyceremia</td>
<td>↑ Insulin sensitivity</td>
<td></td>
</tr>
<tr>
<td>Zhang et al. 2008</td>
<td>Re</td>
<td>40 mg/kg i.p Wistar rats</td>
<td>↓ Fasting glyceremia</td>
<td>↑ Insulin sensitivity</td>
<td></td>
</tr>
<tr>
<td>Kang et al. 2008</td>
<td>Rg3</td>
<td>5,10,20 mg/kg oral STZ- rats</td>
<td>↓ Serum glucose and glycosylated proteins</td>
<td>Inhibit NMDA receptor-mediated nitrosative stress</td>
<td></td>
</tr>
<tr>
<td>Kim et al. 2008</td>
<td>KRG extract</td>
<td>100 mg/kg Male Wistar rats</td>
<td>↓ Serum glucose</td>
<td>↓ Oxidative stress</td>
<td></td>
</tr>
<tr>
<td>Liu et al. 2009</td>
<td>Malonyl-ginsenosides</td>
<td>120 mg/kg STZ-diabetic mice</td>
<td>↓ Fasting glyceremia</td>
<td>Not specified</td>
<td></td>
</tr>
<tr>
<td>Lee et al. 2009</td>
<td>KRG</td>
<td>200 mg/kg OLETF rats</td>
<td>↓ Fasting glucose and HbA1c</td>
<td>↑ Insulin sensitivity</td>
<td></td>
</tr>
<tr>
<td>Yuan et al. 2010</td>
<td>Asian ginseng leaf extract</td>
<td>250-500 mg/kg HFD induced mice</td>
<td>↓ Levels of plasma glucose and insulin</td>
<td>Inhibiting gluconeogenesis and stimulating lipolysis, respectively via AMPK activation</td>
<td></td>
</tr>
<tr>
<td>Yang et al. 2010</td>
<td>Panax notoginseng saponins (PNS); Rb1</td>
<td>PNS: 200mg/kg; Rb1: 60 mg/kg KK-Ay mice</td>
<td>↓ Fasting blood glucose levels; improved glucose tolerance; ↓ serum insulin levels and insulin resistance index</td>
<td>Improving insulin sensitivity in glucose disposal</td>
<td></td>
</tr>
<tr>
<td>Amin et al. 2011</td>
<td>AG aqueous ginseng extract</td>
<td>300 mg/kg i.p STZ induced diabetic rats</td>
<td>Significantly ameliorated the induced hyperglycemia and hypoinsulinemia</td>
<td>Blocking intestinal glucose absorption and inhibiting hepatic G6Pase</td>
<td></td>
</tr>
</tbody>
</table>
Yuan et al. 2011

<table>
<thead>
<tr>
<th>Study</th>
<th>Treatment</th>
<th>Dose</th>
<th>Outcomes</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>31</td>
<td>Pectinase-processed ginseng radix</td>
<td>300 mg/kg</td>
<td>HFD-fed ICR mice</td>
<td>Improved glucose tolerance after glucose challenge, ↓ FBG, FPI, and the insulin resistance index</td>
</tr>
<tr>
<td>32</td>
<td>IH-901, an intestinal metabolite of ginsenosides from Panax ginseng</td>
<td>10, 25 mg/kg</td>
<td>C57BL/KsJ db/db mice</td>
<td>↓ Fasting glucose levels, ↑ plasma insulin levels</td>
</tr>
</tbody>
</table>

Re, Rh2, Rg1, Rb1, Rd, Re, Rg3- conventional nomenclature of ginsenosides; Compound K-protopanaxadiol metabolite; PPD- protopanaxadiols; AG- American Ginseng; KRG- Korean Red Ginseng; ICR- Imprinting Control Region; i.p.- intraperitoneal; i.v.- intravenous; ZDF- Zucker diabetic fatty; STZ- streptozotocin; NMDA- N-methyl-D-aspartic acid; HFD- high fat diet

2.2.6.1.2 Clinical evidence

Prior to the year 2000, scant evidence existed in humans supporting the traditional use of ginseng in diabetes, as only a small group of flawed published studies was available. Of these was a study by Sotaniemi and coworkers\textsuperscript{12}, where they showed that 100 or 200 mg of an unspecified ginseng administered for 8 weeks in 36 subjects with diabetes resulted in significant reductions in HbA1c levels and fasting blood glucose. However, their results were confounded by significant weight losses between the treatment groups, and thus could not be reliable. In another study, Tetsutani et al.\textsuperscript{13} reported that 24 months of KRG extract administration at doses from 3 to 4.5 g reduced HbA1c in 34 people with T2DM compared with controls. But the subject selection, allocation to treatment, statistics, and follow-up of the study were very poorly described. In light of the need for well designed human clinical trials assessing the efficacy of ginseng in diabetes management, our group initiated a clinical testing program where the acute and chronic effects of both AG and KRG were explored, as outlined in Table 4 and Table 5.

Using a 25-g OGTT protocol, a series of five randomized placebo-controlled acute clinical trials were conducted in order to evaluate the efficacy of American ginseng in reducing
postprandial glycemia and its dosing and timing effects in subjects with and without diabetes. Overall, these acute studies showed a significant reduction in PPBG by ~15-22%, and the main findings were 4-fold: (1) AG showed significant improvements in incremental area under the glucose curve (iAUC) by 9-39% (2) doses from 1 to 9 g were equally efficacious; (3) time from 0 to 120 min before the glucose challenge was equally efficacious in diabetic subjects without interaction with their background antihyperglycemic therapy; and (4) only AG administered > 40 min before the OGTT reduced glycemia in nondiabetic subjects. Based on these results, we concluded that AG has the potential to acutely reduce PPBG. The acute promising results seen with our efficacious AG batch were confirmed with a long-term trial using an AG extract with a ginsenoside profile similar to that of AG used in the five acute studies. In an 8-week double-blind, placebo-controlled crossover trial conducted in 24 subjects with T2DM, 1 g of AG extract taken 40 min before each meal (3 g/day) showed significant reductions in FPG and HbA1c compared to placebo. An observable but insignificant increase in insulin was also seen, suggesting a possible improvement in β-cell function. It is noteworthy that these benefits occurred without increasing adverse events suggesting a safety profile for ginseng. Additionally, aiming at exploring whether the glycemia-lowering effect of AG root is batch dependent, we evaluated the effect of 5 root batches, representative of Ontario-grown AG, on PPBG and insulin indices in 12 healthy subjects. On five separate visits, participants were assigned to randomly consume 9 g of AG from 5 farms taken 40 minutes before a 2-hr 75-gram OGTT. A water-control during the 6th and last visit was also administered. Relative to control, we found that the mean of the 5 ginseng treatments reduced peak PPBG by 1.0 mmol/L, glucose IAUC by ~28%, and insulin iAUC by 24%. Our findings suggested that AG root has potential to reduce postprandial glycemia and spare insulin secretion and may complement antidiabetic treatment, without unwanted hypoglycemia. Yet, because of the unexpected null effects observed with ~40% of commercially available, irrespective of their saponin composition, it was concluded that the ginsenosides may not be the sole and best predictors of ginseng’s hypoglycemic activity.
Moreover, an acute study exploring different AG extracts was conducted so as to identify the preparation of AG that is most effective in improving glycemia. Whole root or 30%, 50% and 70% ethanol extracts of AG were administered to 13 NG individuals in a crossover design. The study was unsuccessful in revealing an effect on the iAUC for glucose and insulin; yet, it did show that insulin sensitivity was significantly improved with the 50% ethanol extract. We also explored the acute glycemic effects of KRG in a similar approach where a batch-finding study of various KRG root fractions was conducted followed by a dose-finding study of the most efficacious fraction, both following a double-blind, randomized, within-subject design. In our batch-finding study, 7 healthy subjects received 6 g placebo and KRG-rootlets, -body, and -H$_2$O extract 40 min before a 50-g OGTT. As for the dose-finding study, 12 healthy subjects received 0 g (placebo), 2, 4, and 6 g of the most efficacious root fraction following the same protocol. Results of the batch-finding study showed a wide variation in the ginsenoside profiles across the three root fractions. This variation coincided with differential effects, although the PPD:PPT ratio was irrelevant. A 29% reduction in AUC was observed with KRG-rootlets relative to placebo, while KRG-H$_2$O extract and KRG-body did not show improvements in glycemia. In the dose-finding trial, a significant effect of KRG-rootlets treatment (mean of three doses) but not dose was found. Compared to placebo, a 17% reduction in AUC with the mean of the three doses was observed. Collectively, these two studies demonstrated that 2 g KRG-rootlets was sufficient to achieve reproducible reductions in postprandial glycemia, which was indicated for long-term exploration. In our long-term study, KRG rootlets at a dose of 2g were applied for investigation in a double-blind, randomized, placebo-controlled, crossover fashion. Nineteen subjects with T2DM received 2 g placebo or KRG rootlets 40 min before each meal (6 g/day) for a period of 12 weeks, while maintaining their usual conventional diabetes medications. FPI and 75-g OGTT derived AUC plasma insulin was significantly decreased on the KRG intervention relative to placebo. As well, no significant changes were observed on FPG measures, while the 75-g OGTT derived AUC plasma glucose showed significant reductions. The combined effect was reflected in a similar 33%
increase in both homeostasis model assessment of insulin resistance (HOMA-IR) and the 75g-OGTT derived insulin sensitivity indices on the KRG rootlets treatment relative to placebo. No increasing adverse events or alterations in hepatic, renal, haemostatic, or blood pressure function was seen. We concluded that KRG rootlets can safely improve markers of glycemic control including glucose and insulin beyond conventional treatment.

Despite our numerous findings indicating a promising potential for ginseng in glycemic control, we also reported on the variability of efficacy of Panax ginseng root fractions on PPBG response\textsuperscript{109,110} concluding that standardization of key features of the ginsenoside profile may be responsible for reproducibility and efficacy. This variability has been also shown across different ginseng species. In 2 RCTs involving 22 healthy participants, we found that acute administrations of Asian ginseng at two dose ranges, low; 1,2, and 3g, and high; 3,6, and 9g, demonstrated both null and opposing effects on postprandial indices of glucose and insulin regulation. It was shown that a low total ginsenoside profile along with an altered PPD:PPT ratio (<1), paralleled with no effects of either the doses or the pooled treatments on incremental plasma glucose and insulin concentrations. However, glycemia was found to be significantly higher for the mean of all Asian ginseng doses at the diagnostically and therapeutically relevant two-hr time point when compared to placebo\textsuperscript{30}. In a randomized, double-blind, placebo-controlled crossover study design in 12 healthy subjects, our group also showed that acute administrations (3g) of 8 of the most popular types of Panax and non-Panax species of ginseng, including American, American-wild, Asian, Asian red, Vietnamese, Siberian, Japanese, and Sanchi ginsengs, resulted in variable glycemic effects\textsuperscript{187}. AG lowered, while Asian ginseng, Wild-AG, and Siberian ginseng raised various indices of plasma glucose, and Sanchi ginseng, Asian red ginseng, Japanese ginseng, and Vietnamese ginseng had null effects, all of which were again linked to the PPD:PPT ratio.

Aside from our group, multiple trials from a group in the United Kingdom demonstrated ginseng’s potential in the regulation of blood glucose\textsuperscript{188-190}. In a series of 3 single-dose randomized controlled trials involving >87 healthy subjects, 200 mg and 400 mg of orally-
provided ginseng significantly reduced blood glucose at all three of the post-treatment follow-ups\textsuperscript{188-190}. However, the investigators were unable to find a long-term effect on glucose regulation when 31 non-diabetic participants administered Panax ginseng for 8 weeks, suggesting against the long term use of ginseng in individuals with normal glucose control\textsuperscript{191}. Furthermore, a study by Ma et al.\textsuperscript{192} showed that Panax ginseng root (originating from China) administered at a dose of 2.2 g/day for 4 weeks resulted in significant reductions (~45\%) in HOMA-IR, and FPG (8.1 mmol/L) relative to placebo, supporting an insulin-sensitizing effect of Panax ginseng, which may particularly relate to hepatic insulin sensitivity. Additionally, a report by Reeds et al., showed a neutral effect of orally ingested KRG and ginsenoside Re on β-cell function and insulin sensitivity in overweight and obese individuals with diabetes or IGT. In this study, compared to placebo, neither oral Re (250-500mg/day) nor oral KRG (8g/day) consumed for 30 days resulted in a therapeutic effect on insulin sensitivity or β-cell function\textsuperscript{193}. Finally, four recent trials conducted in different study designs and populations reported both promising and ineffective results for the use of ginseng in diabetes management. In the first study, 8-week administrations of 1500, 2000, or 3000 mg of ginsam, a vinegar extract from Panax ginseng, in subjects with T2DM resulted in significant improvements in HbA1c, and was not associated with any severe adverse events\textsuperscript{194}. In the second study, however, 4.5 g of KRG consumed by 60 subjects with metabolic syndrome for a period of 12 weeks did not show any improvements in FPG measures\textsuperscript{195}. Nevertheless, the study was limited by several factors, suggesting that further long-term investigation with a larger population is warranted. Thirdly, the effect of KRG on insulin sensitivity in 68 healthy, overweight, or obese adults was explored\textsuperscript{196}. It was found that 6 g of KRG rootlets taken daily over a 12 week period led to insignificant improvements in insulin sensitivity, concluding that KRG does not improve the insulin sensitivity of overweight and obese subjects who do not have diabetes or hypertension. Finally, in a 2-week randomized double-blind parallel trial, the effect of fermented red ginseng was assessed in 93 postmenopausal women, where women in the FRG group exhibited significantly decreased levels
of HbA1c, insulin, and homeostatic model assessment of insulin resistance (HOMA-IR)\(^1\) compared to those in the placebo group.

Taken as a whole, and despite mounting clinical evidence in support of an anti-diabetes indication for ginseng, the reproducibility of its safety and efficacy still remains questionable. This emphasizes the need for further exploration and identification of its compositional markers that are responsible for the anti-hyperglycemic effects, such as other unmeasured saponins and nonsaponin fractions. There is also a clear need to establish a basis for standardization that ties the active constitutions of ginseng to its effective glycemic-lowering effects.

**Table 4. Acute and long-term studies conducted in normoglycemic individuals and those with T2DM investigating the dose, time of administration and efficacy of American Ginseng at the Risk Factor Modification Centre, St. Michael’s Hospital (Toronto, Canada)**

<table>
<thead>
<tr>
<th>STUDY</th>
<th>SAMPLE SIZE</th>
<th>TREATMENT</th>
<th>iAUC REDUCTION</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vuksan et al. 2000(^1)</td>
<td>10 NG</td>
<td>3g AG vs. placebo</td>
<td>18%</td>
</tr>
<tr>
<td>Vuksan et al. 2000(^1)</td>
<td>9 T2DM</td>
<td>3g AG vs. placebo</td>
<td>22%</td>
</tr>
<tr>
<td>Vuksan et al. 2000(^1)</td>
<td>10 NG</td>
<td>3, 6 or 9g AG vs. placebo</td>
<td>26, 29 or 39%</td>
</tr>
<tr>
<td>Vuksan et al. 2000(^1)</td>
<td>10 T2DM</td>
<td>3,6 or 9g AG vs. placebo</td>
<td>20, 15 or 16%</td>
</tr>
<tr>
<td>Vuksan et al. 2001(^1)</td>
<td>12 NG</td>
<td>1,2 or 3g AG vs. placebo</td>
<td>14, 15, or 9%</td>
</tr>
<tr>
<td>Dascalu et al. 2007(^1)</td>
<td>12 NG</td>
<td>9g AG (5 farms) vs. placebo</td>
<td>28%</td>
</tr>
<tr>
<td>De Souza 2008(^1)</td>
<td>13 NG</td>
<td>3g whole root, 30%, 50%, 70% AG extract vs. placebo</td>
<td>NS but ↑ ISI with 50% &amp; 70% extract</td>
</tr>
<tr>
<td>Vuksan et al. 2001(^1)</td>
<td>24 T2DM</td>
<td>3g/d AG vs. placebo</td>
<td>10%</td>
</tr>
</tbody>
</table>

NG- normoglycemic; T2DM- Type 2 diabetes mellitus AG- American Ginseng; iAUC- incremental area under the glucose curve; NS- not significant; ISI; insulin sensitivity index
Table 5. Acute and long-term studies conducted in normoglycemic individuals and those with T2DM investigating the dose, different root components and efficacy of Korean Red Ginseng at the Risk Factor Modification Centre, St. Michael’s Hospital (Toronto, Canada).

<table>
<thead>
<tr>
<th>STUDY</th>
<th>SAMPLE SIZE</th>
<th>TREATMENT</th>
<th>iAUC REDUCTION</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sievenpiper et al. 2006¹⁸⁶</td>
<td>7 NG</td>
<td>6g KRG-rootlets, body, H20 extract vs. placebo</td>
<td>29% for KRG rootlets</td>
</tr>
<tr>
<td>Sievenpiper et al. 2006¹⁸⁶</td>
<td>12 NG</td>
<td>2,4,6g KRG vs. placebo</td>
<td>17%</td>
</tr>
<tr>
<td>De Souza et al. 2011¹⁰⁹</td>
<td>13 NG</td>
<td>3g of either KRG body or rootlets vs. placebo</td>
<td>↓ BG iAUC by 27% with KRG body</td>
</tr>
<tr>
<td>Vuksan et al. 2008¹⁵</td>
<td>19 T2DM</td>
<td>6g/d KRG vs. placebo</td>
<td>8-11%</td>
</tr>
</tbody>
</table>

NG- normoglycemic; T2DM- Type 2 Diabetes Mellitus KRG- Korean Red Ginseng; iAUC- incremental area under the glucose curve; NS- not significant; ISI; insulin sensitivity index

2.2.6.2 The effect of ginseng on vascular health

The effect of ginseng on various cardiovascular parameters has been explored. These include BP, endothelial function, cardiac function, and lipid metabolism. Of these, BP has gained much attention, and will only be covered herein. Evidence on its BP health effects from in vitro, animal, and clinical investigations will be discussed.

2.2.6.2.1 Pre-clinical evidence

There has been a sensible amount of basic research focusing on the vascular effects of ginseng, particularly of its postulated active components, ginsenosides, as well as its different types and extracts. Overall, ginseng extract appears to exert a direct vasodilatory effect on isolated blood vessels that may be due to endothelium-dependent release of nitric oxide (NO)²⁰⁰-²⁰². Some of the other cardio-protective mechanisms investigated demonstrate a role of ginsenosides in the blockade of calcium channels²⁰³,²⁰⁴.

Studies conducted earlier where the vascular effects of different ginseng extractions were compared revealed that administration of ether-extracted Korean ginseng showed a
cardiodepressive and a BP lowering effect in anaesthetized dogs, while aqueous extracts generally exerted opposite effects\textsuperscript{205}. This demonstrates that the extraction procedure may play an important role in eliciting different vascular effects, as differences in ginsenoside content become apparent. Additionally, the ginsenoside fraction of KRG administered to anesthetized rats at a dose of (10-100 mg/kg, intravenous) have shown reductions in mean arterial BP in a dose-dependent fashion\textsuperscript{206}, where such effects were found to be associated with vascular endothelial cell-mediated NO release, resulting in the production of cyclic guanosine monophosphate (cGMP), and subsequent relaxation of smooth muscle. Other studies using one-clip Goldblatt hypertensive rat, showed that KRG (1 mg/kg body weight, intravenous) generated an acute and significant reduction in BP (\~30 mmHg) that returned to control levels \~20 minutes after intake\textsuperscript{207,208}. These effects appeared to be related to an increase in NO production secondary to stimulation of NO synthase activity\textsuperscript{207,208}. It is noteworthy that KRG also led to BP reductions in normotensive rats, further supporting the concept of a direct vasodilatating influence that is possibly NO dependent. Though ginseng is characterized by having a complex structure, leading to a composition of a range of different constituents, it is the PPT type ginsenosides that were initially regarded as the agents responsible for improvements in vascular activity\textsuperscript{209}, where greater accumulation of cGMP in rat aortic rings were observed when compared to PPDs\textsuperscript{209}. Amongst all the known and studied PPT type ginsenosides, Rg3 has shown the greatest vasodilatative action\textsuperscript{210}, where they were found to induce endothelium-dependent relaxation in rat thoracic aorta via enhanced release of NO by upregulating the expression of NO synthase\textsuperscript{201}. Collectively, these observations indicate that ginseng exhibits vasodilatative potential presumably via the action of its ginsenoside, though the role of other components cannot be eliminated.
2.2.6.2.2 Clinical evidence

The effects of ginseng on indices of blood pressure activity have been clinically explored across different ginseng species, batches, and preparations in different study populations. Of these, the most compelling evidence for BP lowering effects exists for KRG. An 8-week study by Han et al. revealed that KRG taken at a dose of 4.5g/day in 26 subjects with essential hypertension decreased 24-hr mean systolic BP relative to placebo, though the two intervention phases differed in time length. This effect, nevertheless, was not seen in the normotensive participants who had ‘white coat’ hypertension. The same investigators conducted a trial in 12 healthy normotensive adults, and showed that 610 mg of KRG water extract acutely reduced BP at 45, 60, and 75 minutes post-intervention. Additionally, in a randomized, double-blind, crossover trial conducted by our group in 8 individuals with hypertension, it was found that relative to placebo, 3g of KRG body containing 20mg total ginsenosides acutely lowered systolic BP at 20, 100, and 110-minutes, as well as diastolic BP at 20 and 80-minutes. However, such acute BP lowering effects were not observed with the KRG rootlets which contained total ginsenoside levels of 70mg. As well, in a crossover trial of 17 healthy individuals, we revealed that KRG rootlets as well as an extract of its ginsenoside fraction acutely reduced arterial stiffness, as measured by Al, after 3 hours of treatment, without affecting BP. This however was not seen with the isolated polysaccharide fraction. From this, we deduced that the ginsenoside fraction, and not the polysaccharide, accounted for the beneficial vascular effects of KRG. We also found a significant acute lowering effect of an Rg3-enriched KRG extract on office BP, central BP and central Al, after 3 hours of treatment administration in healthy individuals. This was also seen in another acute trial, where significant reductions in ambulatory systolic and diastolic BP were seen in hypertensive and pre-hypertensive individuals, following administration of an KRG extract with a medium Rg3 dose, rather than a low or high dose.

Despite such promising results, however, two more recent long-term investigations of KRG administered at doses of 3 and 4.5 g/day for 12 weeks failed to demonstrate significant effects
Asian ginseng has also shown blood pressure lowering effects. In a report by Caron et al., *Panax ginseng* extract (Ginsana G115) has shown to acutely reduce blood pressure after 2-hours of administration relative to baseline measures\(^2\).217.

On the other hand, less consistent blood pressure lowering effects were seen with AG compared to KRG. Two studies conducted by our group failed to show beneficial BP reduction effects for AG in subjects with hypertension\(^2\);\(^2\)19, while a more recent long term study conducted in 64 subjects with T2DM and concomitant hypertension revealed that 3g of AG significantly lowered radial Al, and systolic BP when compared to placebo\(^2\)20. A recent study by Yoon et al. demonstrated that chronic ingestion (8-weeks) of 1500, 2000, or 3000 mg of ginsam, a vinegar extract of *Panax ginseng*, did not significantly improve BP measures relative to control\(^1\)94. Taken as a whole, the potential benefit of ginseng as an antihypertensive agent remains unclear. As with other issues related to the therapeutic benefit of ginseng for certain vascular disorders, the type of ginseng may reflect the nature of its potential success or failure.

### 2.2.7 Ginseng safety, adverse events, and drug interactions

The notion that ginseng could elevate BP stems from an early prospective investigation of 133 regular ginseng users\(^2\)21. From this study, a ‘ginseng abuse syndrome’ was introduced, where 22 out of 133 patients presented with symptoms such as hypertension, euphoria, restlessness, nervousness, insomnia, skin eruptions, edema, and diarrhea after long-term ginseng use at variable doses, which averaged a daily intake of 3g, but reached as high as 15g in certain cases. Hypotension was also seen by five subjects. However, numerous caveats were associated with this study as it did not have a control group, and the subjects reported the use of different ginseng species including *P.ginseng* and *P.quinquefolius*. As well, the differential BP effects observed, including hypertension and hypotension, could be largely owed to the varying levels of ginsenosides administered, given the numerous ginseng doses ingested. Based on this
observational study, review papers have advised against the use of ginseng in individuals with hypertension.

On the other hand, clinical studies that have emerged recently demonstrate a neutral or beneficial effect of ginseng on BP. A report by Stavro et al. conducted in hypertensive individuals found no significant adverse effects on BP and renal function when AG was administered at a dose of 3g/day for 12 weeks\textsuperscript{219}, while only two subjects reported symptoms of headache and diarrhea. These neutral effects on BP were also observed with acute administrations of AG\textsuperscript{218}. A systematic global review by Coon and Ernst\textsuperscript{222} exploring the adverse effects of Panax ginseng in clinical trials reported that the incidence of adverse events in experimental groups taking ginseng was similar to that for the placebo groups. The most frequently reported adverse events were headaches, sleep disturbances, and gastrointestinal effects. They concluded that their review of clinical trial data of ginseng mono-preparations indicates that these are rarely associated with adverse effects beyond mild and transient disturbances; multi-component preparations may be associated with some adverse effects, but the degree to which these are attributable to ginseng is not clear.

Additionally, findings from our long term trial on KRG, where both its safety and efficacy were explored, showed no differences between the ginseng-treated and the control groups in frequency and severity of adverse events, blood pressure and measures of renal, hepatic, and haemostatic function\textsuperscript{15}. As for drug-herb interactions, potential interaction of ginseng with drugs such as phenelzine, warfarin and alcohol have been documented, and thus, individuals consuming these products are generally advised against the use of ginseng\textsuperscript{222}. Yet, despite these reports, the WHO continues to endorse ginseng as a safe herb, without known side-effects\textsuperscript{223}. On the contrary, the CDA recommendations do not support use of ginseng in diabetes management, largely owing to the lack of sufficient evidence for its safety and efficacy\textsuperscript{224}. 
2.2.8 Summary of the evidence

Despite ample evidence suggesting an anti-diabetes indication for ginseng, the efficacy of ginseng in improving certain glycemic and metabolic parameters remains questionable. This is further complicated by the high variability in the ginsenoside composition seen across specific ginseng species, sources, and extracts, necessitating the need for better ginseng standardization in order to ensure reproducibility and efficacy. With respect to cardiovascular effects, there is an abundance of in-vitro and animal data that repeatedly show vasoactive efficacy of ginseng. Whole ginseng root, ginseng extract, crude saponins and individual ginsenosides have all been implicated as effective components in improving vascular function possibly through the relaxation of endothelium via increased release of NO. Moreover, a very positive safety profile of ginseng is suggested, where a systematic review of adverse reactions to ginseng concludes that ginseng is well tolerated by the majority of people, and that reported adverse reactions are rare, mild and transient, and their incidence has not been significantly higher than those of placebo.

In conclusion, despite a profusion of published data attempting to characterize ginseng and evaluate its efficacy and mechanisms of action, many pharmacological aspects of ginseng and its components remain unknown. Data from RCTs with adequate safety reporting is scarce and is often accompanied with methodological problems. However, some efficacy trends are apparent and additional research will determine if current knowledge can be replicated and will further elucidate mechanisms of action and components responsible for these effects.
CHAPTER 3

RATIONALE, OBJECTIVES, AND HYPOTHESES
3. RATIONALE, OBJECTIVES, AND HYPOTHESES

3.1 RATIONALE

Ginseng, often described as the ‘king herb’, holds an important position in traditional oriental medicine. Due to its purported health benefits, it is one of the most commonly consumed natural health products (NHPs) worldwide. Mounting evidence from cell culture and animal studies reveal that its principal active component, the ginsenosides, may have beneficial properties in glycemic control. Data from selected randomized controlled trials confirm these benefits, suggesting that it may hold a promising position in diabetes management. However, systematic reviews assessing the efficacy and safety of ginseng as a potential herbal remedy for diabetes reveal inconclusive findings. Some show a lack of convincing evidence for benefit, whereas others report a promising potential in glucose regulation. Due to the inconsistency in the literature, the evidence to support the anti-hyperglycemic efficacy of ginseng is limited. Therefore, in order to provide evidence-based guidance for the inclusion of ginseng in diabetes guidelines, a systematic review and meta-analysis of RCTs assessing the effect of ginseng on glycemic parameters in people with and without diabetes was conducted.

Additionally, varieties of KRG have clinically shown therapeutic potential in the management of diabetes and related cardiovascular complications when used in addition to conventional medication. However, presently, limited clinical evidence exists on the health benefits of the white, non-steamed cultivars of Korean ginseng, KWG. It differs from KRG root in that it is not treated by steaming the fresh white roots, a process that produces organoleptic transformation of the roots to a caramel color and pungent taste. While an enhanced biological action of ginseng has been observed with the steamed KRG versus the non-steamed KWG root, evidence also indicates that the steaming process greatly alters the ginsenoside
profile of ginseng\textsuperscript{25}, and thus, may potentially damage their biological properties. As well, loss of the malonyl type ginsenosides has shown to be associated with high heat applications\textsuperscript{26;27}.

In light of the pre-clinical metabolic benefits seen with KWG\textsuperscript{28;29}, coupled with increased costs of KRG as a result of steaming processing\textsuperscript{25}, KWG may potentially offer a cost-effective natural health alternative for improving certain CVD risks. Given the paucity of clinical data on KWG’s medicinal properties, there is an intriguing need to explore its CVD health benefits including glycemic and vascular markers. Therefore, a randomized, double-blind, placebo-controlled, multiple-crossover trial was further undertaken to investigate the acute postprandial glycemic and vascular effects of KWG in T2DM.

3.2 OBJECTIVES

The overall objective of the thesis work is to evaluate the effects of ginseng (the genus \textit{Panax}) on glycemic and vascular health.

Our specific objectives are:

1) To assess the long term (≥30 days) effects of ginseng (the genus \textit{Panax}) on four parameters of glycemic control, including FBG, FPI, HbA1c, HOMA-IR, in a systematic review and meta-analysis of RCTs in people with and without diabetes.

2) To investigate a dose response relationship between escalating doses of KWG and its therapeutic effect on postprandial glycemic and vascular parameters in an acute, randomized, double-blind, placebo-controlled, multiple-crossover trial in T2DM, so as to establish its therapeutic potential and to serve as pilot data for potential long term exploration.
3.3 HYPOTHESES

The overall hypothesis of the thesis work is that ginseng (the genus *Panax*) will improve glycemic and vascular parameters. More specifically, we hypothesis that:

1) Long term (≥30 days) ginseng (the genus *Panax*) administration will improve glycemic parameters including FBG, HbA1c, FPI, and HOMA-IR compared to control in a systematic review and meta-analysis of RCTs in people with and without diabetes.

2) Use of KWG will be safe, and will lower PPBG iAUC, PPBG, AI, and central and brachial BP relative to control in an acute, randomized, double-blind, placebo-controlled, multiple-crossover trial in T2DM.
CHAPTER 4

THE EFFECT OF GINSENG ON GLYCEMIC CONTROL: A SYSTEMATIC REVIEW AND META-ANALYSIS OF RANDOMIZED CONTROLLED TRIALS
4. THE EFFECT OF GINSENG ON GLYCEMIC CONTROL: A SYSTEMATIC REVIEW AND META-
ANALYSIS OF RANDOMIZED CONTROLLED TRIALS

Running title: Ginseng and Glycemic Control

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Manuscript status: Submitted to PLOS ONE
4.1 ABSTRACT

**Background:** The evidence to support an anti-hyperglycemic effect for ginseng remains unclear despite its widespread use in diabetes management, highlighting the need for higher quality evidence to support its inclusion in clinical practice guidelines.

**Purpose:** To elucidate the effect of ginseng on 4 parameters of glycemic control (FBG, FPI, HbA1c, and HOMA-IR) in a systematic review and meta-analysis of RCTs in people with and without diabetes.

**Data sources:** MEDLINE, EMBASE, CINAHL and the Cochrane Library (through July 3, 2013).

**Study selection:** Randomized controlled trials ≥30 days assessing the glycemic effects of ginseng in people with and without diabetes

**Data extraction:** Treatment differences in FBG, FPI, HbA1c, and HOMA-IR were extracted by 2 independent reviewers. The Heyland Methodological Quality Score and the Cochrane risk of bias tool were used to assess study quality and risk of bias respectively.

**Data synthesis:** Sixteen trials were included, in which 16 FBG (n=770), 10 FPI (n=349), 9 HbA1c (n=264), and 7 HOMA-IR (n=305) comparisons were reported. Ginseng significantly reduced FBG compared to control (MD= -0.31 mmol/L [95%CI: -0.59 to -0.03], P=0.03). People with diabetes demonstrated greater FBG reductions compared to those without diabetes (between-group MD -17.30 mg/dL [95%CI -25.95 to -8.47], P=0.001). Although there was no significant effect on FPI, HbA1c, or HOMA-IR, a priori subgroup analyses did show significant reductions in HbA1c in parallel compared to crossover trials (P=0.01).

**Limitations:** Most trials were short (67% trials<12wks), and included participants with a relatively good glycemic control (median HbA1c non-diabetes=5.4% (36 mmol/mol) [2 trials]; median HbA1c diabetes=7.1% (54 mmol/mol) [7 trials])

**Conclusions:** Ginseng modestly yet significantly improved FBG in people with and without diabetes. These findings support its use as a valuable adjunct to conventional diabetes therapy.
Larger and longer randomized controlled trials using standardized ginseng preparations are warranted to provide better estimates of its anti-diabetic efficacy.

**Clinical trial registration:** ClinicalTrials.gov identifier, NCT01841229

**Key words:** *Panax ginseng, Panax quinquefolius*, systematic review, meta-analysis, diabetes mellitus, glycemic control
4.2 INTRODUCTION

Diabetes is reaching epidemic proportions globally, with rates continually rising in both the developed and developing countries. Despite advances in treatment, long-term diabetes management goals generally remain unmet. Meanwhile, interest in CAM continues to grow, becoming one of the major therapeutic approaches sought by individuals with diabetes. Among the most prevalent herbal CAMs, is AG, which has demonstrated significant promise in the management of T2DM, as acknowledged by the 2002 ADA nutrition recommendations review.

Currently, 13 species of ginseng have been identified, of which Asian ginseng (Panax ginseng) and American ginseng (Panax quinquefolius) are the most extensively used and researched. Its pharmacological activity has been attributed to a group of saponins, also known as ginsenosides.

Several controlled clinical trials using American or Asian ginseng have demonstrated its therapeutic potential for glycemic control. However, systematic reviews of such trials investigating the effect of ginseng on glycemic and metabolic parameters were largely inconclusive, concluding a lack of convincing evidence for benefit or reporting promising results for improving glucose metabolism. A previously conducted meta analysis on a single variety of ginseng, KRG, did not show favorable outcomes in the management of T2DM, but was limited to 4 trials with incompatible study designs. Therefore, our objective was to conduct a systematic review and meta-analysis of RCTs assessing for the first time, the glycemic effects of all species of ginseng (the genus Panax) in people with and without diabetes.

4.3 METHODS

Our meta-analysis followed the Cochrane Handbook for Systematic Reviews of Interventions. We reported our findings according to the Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA) guidelines. The review protocol is available online at ClinicalTrials.gov (registration number, NCT01841229).
4.3.1 Data sources and searches

MEDLINE, EMBASE, CINAHL, and the Cochrane Central Register of RCTs were searched from inception through 3 July 2013, using a comprehensive search strategy (Table 1). Manual searches of the reference lists of all selected and review articles supplemented the electronic search. Abstracts and dissertations were also included with no restriction on language.

4.3.2 Study selection

We included RCTs ≥ 30 days which examined the effect of oral ginseng supplementation (all species of the genus Panax) on four endpoints of glycemic control: FBG, FPI, HbA1c, and HOMA-IR, in individuals with and without diabetes. In order to isolate the effect of ginseng, trials that used ginseng as part of a multi-herbal treatment were excluded. We also excluded trials that lacked an adequate control or did not provide suitable endpoints data.

4.3.3 Data extraction and quality assessment

Data were reviewed and extracted by two independent reviewers (E.S., V.D.) using a standardized pro forma. Relevant data extracted included information on authorship, publication year, study design, follow-up duration, blinding, sample size, subject characteristics, ginseng species, preparation, dose and form, comparator, and funding source. Study quality in included reports was assessed using the Heyland Methodological Quality Score (MQS), where a score ≥8 was considered to be high quality. Evaluation was based on quality of study methods, sample selection and follow-up, and intervention. Risk of bias was subjectively assessed using the Cochrane Risk of Bias tool. Domains of bias explored were sequence generation, allocation concealment, blinding, outcome data, and outcome reporting. Discrepancies were resolved by consensus.

Mean (SD) for baseline values, change from baseline differences, and end differences in FBG, FPI, HbA1c, and HOMA-IR were extracted. Missing SDs were imputed from 95% CI, P
values, $t$ or $F$ statistics using published formulae\textsuperscript{234}. When necessary, end-of-study changes in the treatment and control groups provided by individual trials were used to derive a correlation coefficient between intervention and control group outcomes. These correlations were then pooled using random effect modeling, where the pooled value was used to impute missing SDs\textsuperscript{234;237}. A calculated correlation (0.41) was used only for FPI imputation in paired analysis of crossover trials. Where needed, authors were contacted to request additional data.

### 4.3.4 Data Synthesis and analysis

The mean difference (MD) and 95% CI was the summary outcome measure for all endpoints. Data for FBG, FPI, HbA1c, and HOMA-IR were aggregated using Review Manager (RevMan) software version 5.0.25 (The Nordic Cochrane Centre, The Cochrane Collaboration, Copenhagen, Denmark) for all primary analyses. Data were pooled using random effects models, with inverse variance weighting. Each endpoint was also stratified based on diabetes status. Between-treatment change from baseline differences were used as the primary end points and between-treatment end differences were used when these data were not available. All crossover trials underwent paired analyses\textsuperscript{237}, and the start value of the control was used when baseline values were not reported. In the event of multiple comparator arms within the same study, a weighted average was applied to create a single pairwise comparison\textsuperscript{234}. Inter-study heterogeneity was assessed using the Cochran Q statistic with a significance level set at $P<0.10$, and quantified with the $I^2$ statistic where a value $\geq 50\%$ reflected substantial heterogeneity. Potential sources of heterogeneity were investigated by \textit{a priori} subgroup analyses of ginseng form, preparation, and species, follow-up duration, study quality and design, respective baseline glycemic parameters, and diabetes status. Further post-hoc subgroup analysis was undertaken to investigate possible effect modification by funding source. Meta-regressions were used to assess the significance of subgroup differences. The presence of a linear trend in the data based upon respective baseline glycemic parameters and follow-up duration were studied using continuous
metaregression analyses. Sensitivity analyses were performed to assess any undue influences of individual studies on the overall effect estimates, by systematically removing each individual study from the meta-analysis and recalculating the effect estimate from the remaining studies. Publication bias was examined through visual inspection of funnel plots and quantitatively evaluated by Begg's and Egger's tests. Meta-regressions and assessment of publication bias were performed on STATA 12 (StataCorp, College Station, Texas).

4.4 RESULTS

4.4.1 Search results

Figure 1 shows the flow of the literature. We identified 975 publications, of which 930 were excluded on the basis of title and abstract. Of the 45 potential relevant studies that were retrieved and fully reviewed, 30 were further excluded. Therefore, we included a total of 15 reports, providing data from 16 trials in 770 participants on the following endpoints: FBG (16 trial comparisons, n=770), FPI (10 trial comparisons, n=349), HbA1c (9 trial comparisons n=264), and HOMA-IR (7 trial comparisons n=305) 12;14;15;191-194;238-245

4.4.2 Trial characteristics

Table 2 displays the characteristics of the included trials. Nine (56.3%) trials were conducted in 339 participants with diabetes and 7 (43.7%) in 431 participants without diabetes. Trials conducted on diabetes included subjects with type 1 diabetes mellitus (2 trials), type 2 diabetes mellitus (5 trials), and type 2 diabetes mellitus & pre-diabetes (2 trials). Non-diabetes trials included subjects with essential hypertension (1 trial), pre-diabetes (2 trials), and those that were otherwise healthy (4 trials). The median age of the study population was 51 years (IQR 47.1 – 57.8 years). All but one trial 245 were carried out in outpatient settings. Fifty percent of all trials were conducted in Asia with the remaining conducted in North America and Europe (25%
All trials assessed FBG in participants both with (9 trials; n=339; median baseline FBG: 8.3 mmol/L (IQR 7.8-8.8)) and without (7 trials; n=431 median baseline FBG: 5.5 mmol/L (IQR 5.1-5.8)) diabetes; ten trials assessed FPI in participants both with (7 trials; n=272 median baseline FPI 70.8 pmol/L (IQR 63.5-78.9)) and without (3 trials; n=77 median baseline FPI 74.8 pmol/L (IQR 74.4-86)) diabetes; nine trials assessed HbA1c in participants both with (7 trials; n=235 median baseline HbA1c of 7.1% (IQR 6.5-7.6%)) and without (2 trials; n=29 median baseline HbA1c of 5.4% (IQR 5.4-5.5%)) diabetes; and seven trials assessed HOMA-IR in participants both with (6 trials; n=257 median baseline HOMA-IR of 3.8 (IQR 2.2-5.2)) and without (1 trial; n=48 baseline HOMA-IR=3) diabetes.

Eleven trials (68.8%) used parallel and five (31.3%) used crossover designs. Only two species of ginseng were identified across all trials: *Panax ginseng* (12 trials; 75%) and *Panax quinquefolium* (3 trials; 18.8%); one study, Sotaniemi et al., did not specify ginseng source. Ginseng preparations included whole root/rootlets of Korean red, American, or *Panax ginseng* (4 trials; 25%), and extracts of Korean red, American, or *Panax ginseng* (10 trials; 62.5%). Two trials (12.5%) did not specify method of preparation. All the trials used encapsulated powder forms of ginseng as the intervention. Thirteen trials (81.2%) used a placebo as the comparator, 2 (12.5%) used a control group that did not receive ginseng, and 1 (6.25%) used fermented soybean. The median follow-up was 8 weeks (IQR 8-12).

Eleven trials (68.8%) were found to be of high quality (MQS≥8). The median MQS among the available trials was 8 (IQR 7-10) (Table 3). Of those trials which received low scores, the elements contributing to the low scores were poor description of randomization and treatment protocol, nonconsecutive or poorly described patient selection, high drop-out rates, and the absence of an intention-to-treat analysis. The Cochrane Risk of Bias Tool showed that 9 trials (56.3%) had unclear risk of bias and 7 trials (43.7%) had low risk of bias for sequence generation. Six trials (37.5%) had unclear risk of bias and 10 trials (62.5%) had low risk of bias.
for allocation concealment. Two trials (12.5%) had unclear risk of bias, 12 trials (75%) had low risk of bias, and 2 trials (12.5%) had high risk of bias for blinding. Fourteen trials (87.5%) were scored with low risk of bias, and 2 trials (12.5%) with high risk of bias for incomplete outcome data. Ten trials (62.5%) scored unclear risk, and 6 (37.5%) scored low risk of bias for selective outcome reporting (Table 4). Eight (50%) studies reported funding from agency sources, four (25%) reported industry support, one (6.3%) reported agency-industry support, and three (18.7%) did not specify.

4.4.3 Effect on Fasting Blood Glucose:

Figure 2 shows the effect of ginseng supplementation on FBG. Overall, a significant FBG reduction was observed (MD -0.31 mmol/L [95%CI -0.59 to -0.03], P=0.03). However, significant evidence of inter-study heterogeneity was seen in the overall analysis (I²=89%; P <0.001) and in the analyses in individuals both with (I²=72%; P <0.001) and without (I²=63%; P=0.01) diabetes.

Sensitivity analyses of systematically removing individual trials showed that removal of five trials individually demonstrating benefit for FBG \textsuperscript{12;14;192;238;240} led to a loss of significance in the overall effect (MD= -0.16 mmol/L; P=0.14, MD= -0.27 mmol/L; P=0.06, MD= -0.28 mmol/L; P=0.06, MD= -0.28 mmol/L; P=0.05, MD= -0.23 mmol/L; P=0.1 respectively).

A priori subgroup analyses revealed that the FBG-lowering effects of ginseng were only modified by the diabetes status of the individuals (between-group MD -0.96 mmol/L [95%CI -1.44 to -0.47], P=0.001). Studies conducted in individuals with diabetes demonstrated a MD of -0.84 mmol/L [95%CI -1.22 to -0.46], whereas those conducted in individuals without diabetes showed a MD of 0.11 mmol/L [95%CI -0.19 to 0.42] (Figure 3).

Continuous meta-regression analyses revealed a linear association between baseline FBG and treatment differences in FBG (β -0.26 mmol/L [95%CI -0.40 to -0.13] per 1 mmol/L,
Heterogeneity remained significant, and could not be explained away by any of the subgroup analyses.

4.4.4 Effect on Fasting Plasma Insulin:

Figure 4 shows the effect of ginseng supplementation on FPI. Overall, no significant difference in FPI levels were observed (MD 0.16 pmol/L [95%CI -5.04 to 5.37], P=0.95), along with no evidence of inter-study heterogeneity (I²=23%; P=0.23). No difference in effect estimate was identified by diabetes status. Sensitivity analyses did not alter the direction or significance of effect estimates nor modify heterogeneity; and a priori subgroup analyses did not reveal significant effect modification by any subgroup under both dichotomous and continuous models (Figure 5 and Table 5).

4.4.5 Effect on Glycated Hemoglobin:

Figure 6 shows the effect of ginseng supplementation on HbA1c. Ginseng supplementation did not change HbA1c levels in the overall analysis (MD -0.0005% [95%CI -0.005% to 0.004%], P=0.82), nor in the analyses by diabetes status. Nevertheless, significant inter-study heterogeneity was observed in the overall analysis (I²=62%, P=0.007) and in the analyses in participants with diabetes (I²=72%, P=0.002).

In our sensitivity analyses, the systematic removal of 2 individual trials resulted in a similar overall borderline significant effect, both demonstrating a MD of -0.15% [95% Cl -0.29% to -0.001%], P=0.05. The overall heterogeneity in the results was not affected by the individual removal of any studies.

A priori subgroup analyses showed that reductions in HbA1c were only modified by study design (MD 0.22% [95% CI 0.06% to 0.37%], P=0.01). Crossover trials showed a MD of -0.0003% [95% CI -0.004% to 0.003%], whereas parallel trials demonstrated a MD of -0.22% [95% CI -0.37% to -0.06%] (Figure 7). No linear associations between baseline HbA1c or
follow-up duration and reductions in HbA1c were found by continuous meta-regression analyses (Table 5).

4.4.6 Effect on Homeostasis model assessment of insulin resistance:

Figure 8 shows the effect of ginseng supplementation on HOMA-IR. Ginseng supplementation did not affect HOMA-IR levels in the overall analysis (MD 0.0009 [95% CI -0.59 to 0.59], P=1.00), nor in analyses by diabetes. Although, significant inter-study heterogeneity was detected in the overall analysis ($I^2=81\%, \ P<0.001$) and in the analyses in participants with diabetes ($I^2=83\%, \ P<0.001$).

Sensitivity analyses did not alter the direction or significance of effect estimates nor modify heterogeneity; and a priori subgroup analyses did not reveal significant effect modification by any subgroup under both dichotomous and continuous models. (Figure 9 and Table 5).

4.4.7 Publication Bias

Visual inspection of funnel plots suggested asymmetry in the FBG, FPI, and HbA1c analyses, with tendencies for the publication of small trials favoring ginseng for FBG and HbA1c, and favoring comparator for FPI. However, this was not confirmed by either Egger’s or Begg’s tests (Figure 10).

4.5 DISCUSSION

An aggregate analyses of 16 RCTs in 770 participants showed that ginseng supplementation significantly lowered FBG. Ginseng intakes decreased FBG by 0.31 mmol/L (95% CI: -0.59 to -0.03, P=0.03) following duration of ≥1 month, in people with and without
diabetes. Although there was no significant overall effect of ginseng on FPI, HbA1c, or HOMA-IR, \textit{a priori} subgroup analyses did show a significant HbA1c benefit in parallel trials compared to crossover trials. Greater reductions in FBG were also observed in people with diabetes than those without diabetes.

Our findings add to those from previous systematic reviews in this area. Two recent systematic reviews assessing the efficacy and safety of ginseng reported promising, but inconclusive evidence for its application in moderating glucose metabolism\textsuperscript{18,19}. However, another earlier systematic review and meta-analysis failed to show a FBG-lowering effect of ginseng supplementation\textsuperscript{20}. One reason for these inconsistencies may relate to differences in their eligibility criteria. Whereas the earlier systematic review included only RCTs that investigated the effect of KRG in subjects with T2DM with treatment durations of at least 12 weeks, we included trials which investigated the effect of any ginseng species in people with or without diabetes over at least 4 weeks. Despite having more inclusive criteria for ginseng species, diabetes status, and follow-up, we did not find any significant effect modification by any of these criteria with the exception of diabetes status.

Diabetes status partially explained the heterogeneity in the overall analysis for FBG. Participants with diabetes had a greater reduction in FBG than participants without diabetes. In line with this subgroup effect, our continuous meta-regression analyses showed that increases in baseline FBG were linearly associated with FBG reductions on the ginseng intervention, further supporting the notion that ginseng supplementation may generate a greater benefit in individuals with higher FBG levels.

It is unclear why the improvements in HbA1c were restricted to parallel trials only. One explanation may be an inadequate washout period between treatments in crossover designs, as it has been shown that ginseng metabolites may remain in the body for up to 10 weeks after
treatment has ended. As all of the crossover trial washouts were <10 weeks (range 27–42 days), confounding from carryover effects remains a strong possibility. Another explanation may relate to the glycemic control of the participants. It is a well understood phenomenon that the higher baseline HbA1c levels, the greater the fall with anti-hyperglycemic agents. As most of the trials included participants with relatively good glycemic control in people with diabetes (median HbA1c 7.1%), it is possible that this resulted in blunted effects. The analysis also included people without diabetes (22% of trials) with optimal control (median HbA1c 5.4%) in whom a significant reduction in their HbA1c levels is unlikely to be observed. Finally, given the evidence of a half life for HbA1c reductions of ~35 days, the short duration of the majority of included trials (6 of 9 trials that assessed HbA1c had <12 weeks follow-up) may have also underestimated the true HbA1c reductions.

The current results do not support the majority of evidence from animal and in vitro data that suggest a potential of ginseng to increase insulin sensitivity and/or secretion. Nevertheless, meta-regression by ginseng species found greater FPI reductions for Panax ginseng relative to Panax quinquefolium, although the between-subgroup difference was not significant (MD = -13.41 pmol/L ([95%CI: -28.19 to 1.37], P=0.07). These observations are line with findings from previous clinical trials supporting both the proposed insulin sensitizing and insulin secreting mechanisms for Panax ginseng and Panax quinquefolium respectively in the amelioration of hyperglycemia in T2DM.

The mechanisms underlying ginseng's hypoglycemic activity remain unclear. A growing database of cell culture and animal studies indicate that ginseng may alleviate hyperglycemia by enhancing pancreatic β-cell function and reducing insulin resistance. Data from these studies support four possible modes of anti-diabetic action: modulation of (1) glucose absorption, (2) insulin secretion and binding, (3) glucose transport, and/or (4) glucose disposal.

Collectively,
these investigations offer preliminary but plausible explanations for the anti-diabetic potency of
the two ginseng species in the clinical trials of this meta-analysis.

Assessment of the safety and tolerability of ginseng was not possible in this systematic
review and meta-analysis, as only 4 of the 16 trials reported safety parameters among which a
consistent safety parameter did not exist\textsuperscript{14,15,194,243}. Markers used in evaluating ginseng’s safety
included hepatic, renal, haemostatic, BP function, comprehensive blood tests, and number of
adverse events, none of which reported any difference in adverse events relative to the control.
This parallels findings of several systematic reviews investigating the efficacy and safety of
ginseng, where it was concluded that while its efficacy remains questionable, it appears to be
generally safe\textsuperscript{18,249,251}. Due to the nature of the intervention used in the trials, we could not
eliminate the possibility of an effect modification by source of funding. Hence, following a post
hoc analysis, no effect of funding source was found for any of the endpoints.

4.5.1 Limitations

Several limitations of this systematic review and meta-analysis should be acknowledged.
First, although undertaking a dose-response analysis was specified \textit{a priori}, it was not evaluated
in the present work due to the variation in ginseng preparations administered (whole root/rootlets
of Korean red/Panax/American ginseng vs. often uncharacterized extracts of these varieties),
precluding calculation of ginseng dose equivalents. Second, the high variability in ginsenoside
composition along with poor standardization of the ginsenoside profile continues to add
complexity to the assessment of ginseng’s glycemic benefits, as it has been shown that the anti-
hyperglycemic efficacy of ginseng varies across species and is correlated to its ginsenoside
composition\textsuperscript{252}. To date, the optimal ratio of the most prominent bioactive components of ginseng,
the ginsenosides, needed to ensure reproducible glucose lowering effects and product quality has
not been fully determined, necessitating a demand for better ginseng standardization. Third,
information on the ginsenoside profile was not given by most of the trials, complicating the corroboration of a corresponding ginsenoside profile across studies that demonstrated effective findings. Finally, publication bias remains a concern as our funnel plot analyses demonstrated evidence of asymmetry favoring small studies with FBG and HbA1c reducing effects, though this was neither confirmed by Egger’s nor by Begg’s test.

4.5.2 Conclusions

In conclusion, aggregate data analyses of controlled clinical trials show evidence for a modest yet significant benefit of ginseng in improving FBG in people with and without diabetes. Given the importance of FBG as a marker of diabetes onset and management, these findings support the use of ginseng as a valuable adjunct to conventional diabetes therapy. Although ginseng did show advantages for HbA1c in parallel trials, the overall lack of an effect on HbA1c and persistent unexplained heterogeneity among the effect estimates from the available trials creates some uncertainty as to the long-term benefits of ginseng supplementation on glycemic control. The uncertainty points to several methodological limitations including the short duration of the trials, the well controlled glycemia of participants at baseline, and the use of unstandardized ginseng preparations with potentially varying potencies. To provide more precise estimates of ginseng’s long term effectiveness, longer term, large scale, RCTs of the effect of ginseng preparations on HbA1c are warranted. Finally, given the promising effects of ginseng on FBG demonstrated herein, further research in this area is sensible, with a particular focus in investigating its potential glycemic-lowering components such as the unexamined non-saponins.

4.6 ACKNOWLEDGMENTS

We would like to thank Ms. Evelyn Wong and Ms. Shana Kim for providing assistance with the translations of the non-English articles.
4.7 AUTHORS CONTRIBUTION

E.S. was responsible for developing the search strategy, conducting the search, performing data analysis and interpretation, drafting, revising, and finalizing the manuscript. V.V was responsible for conception and design of the project, analysis and interpretation of data, critical revision of the manuscript, and supervision. V.V had full access to all the data in the study and had final responsibility for the decision to submit for publication. J.L.S. was responsible for conception and design of the project, analysis and interpretation of data, and critical revision of the manuscript. D.J.A.J. assisted in the design of the project and critical revision of the manuscript. R.J.d.S. was responsible for interpretation of data, critical revision of manuscript, and statistical analysis. A.C., V.H., and V.H.J. assisted in the search, extraction of data, data interpretation, and critical revision of the manuscript. V.D. assisted in the extraction of study characteristics and data from each included study and critical revision of the manuscript. S.B.M. and E.J. assisted in the search, extraction of data, data interpretation, and critical revision of the manuscript. All authors approved the final manuscript for publication and agreed to be accountable for all aspects of the work.

4.8 CONFLICTS OF INTERST/DISCLOSURE

E.S. received funding from the Embassy of the State of Kuwait, Kuwait University office. V.V. is a holder of an American (No. 7,326,404 B2) and Canadian (No. 2,410,556) patent for use of viscous fibre blend in diabetes, metabolic syndrome and cholesterol lowering; currently holds grant support for ginseng research from the Canadian Diabetes Association, Canada and the National Institute of Horticultural & Herbal Science, RDA, Korea, and is a part owner of the Glycemic Index Laboratories, Inc. a contract research organization. JLS has received research support from the Canadian Institutes of health Research (CIHR), Calorie Control Council, The Coca-Cola Company (investigator initiated, unrestricted grant), Pulse Canada, and The International Tree Nut Council Nutrition Research & Education Foundation. He has received travel funding, speaker fees, and/or honoraria from the American Heart Association (AHA), American College of
Physicians (ACP), American Society for Nutrition (ASN), National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) of the National Institutes of Health (NIH), Canadian Diabetes Association (CDA), Canadian Nutrition Society (CNS), Calorie Control Council, Diabetes and Nutrition Study Group (DNSG) of the European Association for the Study of Diabetes (EASD), International Life Sciences Institute (ILSI) North America, International Life Sciences Institute (ILSI) Brazil, Abbott Laboratories, Pulse Canada, Dr. Pepper Snapple Group, and The Coca-Cola Company. He is on the Clinical Practice Guidelines Expert Committee for Nutrition Therapy of both the Canadian Diabetes Association (CDA) and European Association for the study of Diabetes (EASD), as well as being on the American Society for Nutrition (ASN) writing panel for a scientific statement on the metabolic and nutritional effects of fructose, sucrose and high fructose corn syrup. He is an unpaid scientific advisor for the International Life Science Institute (ILSI) North America, Food, Nutrition, and Safety Program (FNSP). His wife is an employee of Unilever Canada. DJAJ has received consultant fees, honoraria, travel funding, or research support from or served on the scientific advisory board for the CIHR, Canadian Foundation for Innovation (CFI), Ontario Research Fund (ORF), and Advanced Foods and Material Network (AFMNet) Calorie Control Council, The Coca-Cola Company (investigator initiated, unrestricted), Barilla, Solae, Unilever, Hain Celestial, Loblaw Supermarkets, Inc., Sanitarium Company, Herbalife International, Pacific Health Laboratories, Inc., Metagenics/MetaProteomics, Bayer Consumer Care, Oldways Preservation Trust, The International Tree Nut Council Nutrition Research & Education, The Peanut Institute, Procter and Gamble Technical Centre Limited, Griffin Hospital for the development of the NuVal System, Soy Advisory Board of Dean Foods, Alpro Soy Foundation, Nutritional Fundamentals for Health, Pacific Health Laboratories, Kellogg’s, Quaker Oats, The Coca-Cola Sugar Advisory Board, Pepsi Company, Agrifoods and Agriculture Canada (AAFC), Canadian Agriculture Policy Institute (CAPI), The Almond Board of California, The California Strawberry Commission, Orafti, the Canola and Flax Councils of Canada, Pulse Canada, the Saskatchewan Pulse Growers, and Abbott Laboratories. RJdS has received research support from the Canadian
Institutes of Health Research (CIHR), Calorie Control Council, and The Coca-Cola Company (investigator initiated, unrestricted). He has served as an external resource person to the World Health Organization's (WHO) Nutrition Guidelines Advisory Group (NUGAG), and was the lead author of a systematic review and meta-analysis commissioned by the WHO of trans fatty acids and health outcomes. The WHO paid for his travel and accommodation to attend the 5th NUGAG Meeting in Hangzhou, China (4-7 Mar, 2013). VH has received research support from the Ontario Graduate Scholarship (OGS) and the Canadian Institutes of Health Research (CIHR). S.B.M., A.C., V.H.J., V.D., and E.J. have no declared conflicts of interest related to this paper.
4.9 FIGURES

All reports identified through database searching: 975
- MEDLINE (1946 to July 3, 2013): 140
- EMBASE (1947 to July 3, 2013): 720
- CINAHL (1985 to July 3, 2013): 100
- The Cochrane Library (to July 3, 2013): 11
- Manual Searches: 4

Reports excluded on basis of title and/or abstract: 930
- Not human studies: 230
- Acute or short-term reports: 21
- Duplicate reports: 202
- Editorials & commentaries: 9
- Case study reports: 4
- Observational reports: 30
- Reports with no ginseng administration: 19
- Review reports: 391
- Systematic reviews and meta-analyses: 23
- Not a clinical trial: 1

Reports reviewed in full: 45

Reports excluded: 30
- No suitable endpoints: 17
- No suitable control: 13

15 Reports (16 trials) included in the meta-analysis:
- 16 trials assessed ginseng's effect on FBG (n=770)
- 10 trials assessed ginseng’s effects on FPI (n=349)
- 9 trials assessed ginseng’s effect on HbA1c (n=264)
- 7 trials assessed ginseng’s effect on HOMA-IR (n=305)

Figure 1. Flow of the literature search for the effect of ginseng on glycemic outcomes (fasting blood glucose, fasting plasma insulin, glycated hemoglobin, and homeostasis model assessment of insulin resistance)
**Figure 2.** Forest plots of controlled clinical trials investigating the effect of ginseng on fasting blood glucose. The diamond represents a pooled effect estimate. Paired analyses were applied to all crossover trials. Data are mean differences (MD) with 95% CI. P values are for Generic Inverse Variance random effects models. Inter-study heterogeneity was tested by the Cochran Q statistic at a significance level of \( P < 0.10 \) and quantified by the I\(^2\) statistic, where I\(^2\) \( \geq 50\% \) is considered to be evidence of substantial heterogeneity.
<table>
<thead>
<tr>
<th>Subgroup</th>
<th>Level</th>
<th>No. trials</th>
<th>N</th>
<th>Mean difference [95% CI] in FBG, mmol/L</th>
<th>Residual I²</th>
<th>P-value</th>
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<td></td>
<td>Within subgroups [95% CI]</td>
<td>Between subgroups [95% CI]</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>16</td>
<td>770</td>
<td>-0.31(-0.59 to-0.03)</td>
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<tr>
<td>Ginseng Form</td>
<td>Powder</td>
<td>16</td>
<td>770</td>
<td>-0.31(-0.59 to-0.03)</td>
<td>-</td>
<td>-</td>
</tr>
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<td>Other</td>
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<tr>
<td>Follow-up</td>
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<td>M2S</td>
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<td>0.60(-0.12 to 1.31)</td>
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<td>≥8</td>
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<td>583</td>
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<td>-0.20(-1.09 to 0.69)</td>
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<td>670</td>
<td>-0.31(-0.77 to 0.15)</td>
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<td>≤5.6 mmol/L</td>
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<td>304</td>
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<td>-0.64(-1.34 to 0.06)</td>
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<td>&gt;5.6 mmol/L</td>
<td>11</td>
<td>466</td>
<td>-0.60(-1.04 to -0.15)</td>
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<td>Species</td>
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<td>133</td>
<td>-0.96(-1.84 to -0.07)</td>
<td>0.74(-0.23 to 1.71)</td>
<td>88.7% 0.12</td>
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<td>601</td>
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<td>Diabetes</td>
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<td>339</td>
<td>-0.84(-1.22 to -0.46)</td>
<td>-0.96(-1.44 to -0.47)</td>
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<td>No</td>
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<td>431</td>
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<td>197</td>
<td>-0.24(-0.89 to 0.42)</td>
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**Figure 3.** Forest plots of subgroup analyses investigating the effect of ginseng on fasting blood glucose. Data are mean differences (MD) with 95% CI. N represents the number of participants in each subgroup. Between subgroup differences were analyzed using meta-regression, with the residual I² reported as a percent value and significance as a P-value, with P <0.05 as significant*. All subgroups were pre-specified as a priori except for funding (post-hoc).
**Figure 4.** Forest plots of controlled clinical trials investigating the effect of ginseng on fasting plasma insulin. The diamond represents a pooled effect estimate. Paired analyses were applied to all crossover trials. Data are mean differences (MD) with 95% CI. P values are for Generic Inverse Variance random effects models. Inter-study heterogeneity was tested by the Cochran Q statistic at a significance level of $P < 0.10$ and quantified by the $I^2$ statistic, where $I^2 \geq 50\%$ is considered to be evidence of substantial heterogeneity.
### Figure 5. Forest plots of subgroup analyses investigating the effect of ginseng on fasting plasma insulin. Data are mean differences (MD) with 95% CI. N represents the number of participants in each subgroup. Between subgroup differences were analyzed using meta-regression, with the residual $I^2$ reported as a percent value and significance as a $P$-value, with $P < 0.05$ as significant*. All subgroups were pre-specified as a priori except for funding (post-hoc).
Figure 6. Forest plots of controlled clinical trials investigating the effect of ginseng on glycated hemoglobin. The diamond represents a pooled effect estimate. Paired analyses were applied to all crossover trial. Data are mean differences (MD) with 95% CI. P values are for Generic Inverse Variance random effects models. Inter-study heterogeneity was tested by the Cochran Q statistic at a significance level of $P < 0.10$ and quantified by the $I^2$ statistic, where $I^2 \geq 50$% is considered to be evidence of substantial heterogeneity.
<table>
<thead>
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<th>Subgroup Level</th>
<th>No. trials</th>
<th>N</th>
<th>Mean difference [95% CI] in HbA1c, %</th>
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<th>P-value</th>
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<td>Total</td>
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<td>264</td>
<td>-0.0005 [-0.005 to 0.004]</td>
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<td>Ginseng Form</td>
<td>Powder</td>
<td>9</td>
<td>264</td>
<td>-0.0003 [-0.003 to 0.004]</td>
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<td></td>
<td>Other</td>
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<td>-</td>
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<td>Ginseng Prep</td>
<td>Whole root/rootlets</td>
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<td>176</td>
<td>-0.10 [-0.28 to 0.07]</td>
<td>-0.11 [-0.48 to 0.24]</td>
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<td>Follow-up</td>
<td>&lt;8 weeks</td>
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<td>15</td>
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<td>≥8 weeks</td>
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<td>249</td>
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<td>MQS &lt;8</td>
<td>3</td>
<td>98</td>
<td>-0.11 [-0.39 to 0.18]</td>
<td>-0.02 [-0.37 to 0.33]</td>
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<td>≥8</td>
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<td>166</td>
<td>-0.12 [-0.34 to 0.09]</td>
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<td>0.22 [0.06 to 0.37]</td>
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<td>Parallel</td>
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<td>-0.22 [-0.37 to -0.06]</td>
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<tr>
<td>Baseline &lt;6.5%</td>
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<td>165</td>
<td>-0.04 [-0.28 to 0.20]</td>
<td>-0.15 [-0.48 to 0.18]</td>
<td>52.3% 0.32</td>
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<tr>
<td></td>
<td>&gt;6.5%</td>
<td>5</td>
<td>99</td>
<td>-0.19 [-0.42 to 0.04]</td>
<td></td>
</tr>
<tr>
<td>Species Panax quinquefolius</td>
<td>1</td>
<td>24</td>
<td>-0.002 [-0.32 to 0.32]</td>
<td>-0.15 [-0.52 to 0.22]</td>
<td>53.0% 0.38</td>
</tr>
<tr>
<td></td>
<td>Panax ginseng</td>
<td>7</td>
<td>204</td>
<td>-0.15 [-0.34 to 0.03]</td>
<td></td>
</tr>
<tr>
<td>Diabetes Yes</td>
<td>7</td>
<td>235</td>
<td>-0.0004 [-0.006 to 0.005]</td>
<td>-0.04 [-1.06 to 0.97]</td>
<td>66.9% 0.92</td>
</tr>
<tr>
<td></td>
<td>No</td>
<td>2</td>
<td>29</td>
<td>0.04 [0.97 to 1.06]</td>
<td></td>
</tr>
<tr>
<td>Funding Agency</td>
<td>3</td>
<td>7</td>
<td>10</td>
<td>-0.10 [-0.36 to 0.16]</td>
<td>-0.01 [-0.40 to 0.37]</td>
</tr>
<tr>
<td></td>
<td>Industry</td>
<td>4</td>
<td>123</td>
<td>-0.11 [-0.40 to 0.17]</td>
<td></td>
</tr>
</tbody>
</table>

**Figure 7.** Forest plots of subgroup analyses investigating the effect of ginseng on glycated hemoglobin. Data are mean differences (MD) with 95% CI. N represents the number of participants in each subgroup. Between subgroup differences were analyzed using meta-regression, with the residual $I^2$ reported as a percent value and significance as a P-value, with $P < 0.05$ as significant*. All subgroups were pre-specified as a priori except for funding (post-hoc).
Figure 8. Forest plots of controlled clinical trials investigating the effect of ginseng on homeostasis model assessment of insulin resistance. The diamond represents a pooled effect estimate. Paired analyses were applied to all crossover trials. Data are mean differences (MD) with 95% CI. P values are for Generic Inverse Variance random effects models. Inter-study heterogeneity was tested by the Cochran Q statistic at a significance level of $P < 0.10$ and quantified by the $I^2$ statistic, where $I^2 \geq 50\%$ is considered to be evidence of substantial heterogeneity.
<table>
<thead>
<tr>
<th>Subgroup</th>
<th>Level</th>
<th>No. trials</th>
<th>N</th>
<th>Mean difference [95% CI] in HOMA-IR</th>
<th>Residual I²</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Within subgroups [95% CI]</td>
<td>Between subgroups [95% CI]</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>7</td>
<td>305</td>
<td>0 (-0.59 to 0.59)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Ginseng Form</td>
<td>Powder</td>
<td>7</td>
<td>305</td>
<td>0 (-0.59 to 0.59)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Other</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Ginseng Preparation</td>
<td>Whole root/rootlets</td>
<td>3</td>
<td>123</td>
<td>0.79 (-0.09 to 1.67)</td>
<td>-0.05 (-1.87 to -0.04)</td>
<td>53.6%</td>
</tr>
<tr>
<td></td>
<td>Extract</td>
<td>3</td>
<td>180</td>
<td>-0.16 (-0.41 to 0.09)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Follow-up</td>
<td>&lt;8 weeks</td>
<td>2</td>
<td>104</td>
<td>-0.69 (-2.40 to 1.02)</td>
<td>0.92 (-1.11 to 2.95)</td>
<td>81.6%</td>
</tr>
<tr>
<td></td>
<td>≥8 weeks</td>
<td>3</td>
<td>201</td>
<td>0.24 (-0.86 to 1.32)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>MQS</td>
<td>&lt;8</td>
<td>2</td>
<td>62</td>
<td>-0.36 (-2.11 to 1.40)</td>
<td>0.47 (-1.61 to 2.56)</td>
<td>81.6%</td>
</tr>
<tr>
<td></td>
<td>≥8</td>
<td>5</td>
<td>243</td>
<td>0.11 (-1.02 to 1.25)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Design</td>
<td>Crossover</td>
<td>3</td>
<td>63</td>
<td>0.23 (-1.36 to 1.81)</td>
<td>0.38 (-1.39 to 2.35)</td>
<td>63.9%</td>
</tr>
<tr>
<td></td>
<td>Parallel</td>
<td>4</td>
<td>242</td>
<td>-0.15 (-1.32 to 1.01)</td>
<td>-</td>
<td>-</td>
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<tr>
<td>Baseline</td>
<td>≤3.1</td>
<td>4</td>
<td>189</td>
<td>0.18 (-0.94 to 1.30)</td>
<td>-0.72 (-2.78 to 1.34)</td>
<td>83.7%</td>
</tr>
<tr>
<td></td>
<td>&gt;3.1</td>
<td>3</td>
<td>116</td>
<td>-0.34 (-2.27 to 1.20)</td>
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<td>-</td>
</tr>
<tr>
<td>Species</td>
<td>Panax quinquefolius</td>
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<td>108</td>
<td>-0.05 (-1.91 to 1.80)</td>
<td>0.03 (-2.20 to 2.26)</td>
<td>82.4%</td>
</tr>
<tr>
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<td>Panax ginseng</td>
<td>5</td>
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<td>-0.02 (-1.27 to 1.22)</td>
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<td>-</td>
</tr>
<tr>
<td>Diabetes</td>
<td>Yes</td>
<td>6</td>
<td>257</td>
<td>-0.12 (-1.01 to 0.78)</td>
<td>-1.72 (-5.56 to 2.12)</td>
<td>82.7%</td>
</tr>
<tr>
<td></td>
<td>No</td>
<td>1</td>
<td>48</td>
<td>1.60 (-2.13 to 5.33)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Funding</td>
<td>Agency</td>
<td>5</td>
<td>209</td>
<td>-0.04 (-1.20 to 1.13)</td>
<td>0.03 (-2.38 to 2.45)</td>
<td>83.9%</td>
</tr>
<tr>
<td></td>
<td>Industry</td>
<td>2</td>
<td>96</td>
<td>-0.004 (-2.12 to 2.12)</td>
<td>-</td>
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</tr>
</tbody>
</table>

**Figure 9.** Forest plots of subgroup analyses investigating the effect of ginseng on homeostasis model assessment of insulin resistance. Data are mean differences (MD) with 95% CI. N represents the number of participants in each subgroup. Between subgroup differences were analyzed using meta-regression, with the residual I² reported as a percent value and significance as a P-value, with P < 0.05 as significant*. All subgroups were pre-specified as a priori except for funding (post-hoc).
Figure 10. Funnel plot assessing publication bias and effect of small study effects in clinical trials investigating the effects of ginseng on A. Fasting blood glucose, B. Fasting plasma insulin, C. Glycated hemoglobin, and D. Homeostasis model assessment of insulin resistance. The dashed lines represent the pooled effect estimate expressed as a mean difference (MD). The diamonds represent within subgroup MD, and the horizontal lines represent standard errors of the MD.
Table 1. Search strategy for studies assessing the effect of ginseng on glycemic control in randomized controlled trials*

<table>
<thead>
<tr>
<th>Database</th>
<th>Search period</th>
<th>Search</th>
</tr>
</thead>
<tbody>
<tr>
<td>MEDLINE</td>
<td>1946 to week 1 of July 2013</td>
<td>1. exp Panax/ or ginseng.mp.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2. exp Hemoglobin A, Glycosylated/ or exp Fructosamine/ or fructosamine.mp. or glycemia.mp or exp Hyperglycemia/ or fructosamine.mp. or diabetes mellitus.mp. or exp Insulin/ or dysoxemia.mp. or hyperinsulin.mp. or hyperinsulin*.mp. or exp Diabetes Mellitus, Type1/ or type 1 diabetes.mp. or exp Diabetes Mellitus, Type2/ or type 2 diabetes.mp. or glib albumin.mp. or exp Diabetes, Gestational/ or gestational diabetes.mp. or exp Prediabetic State/ or prediabetes.mp. or HBA1C.mp.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>3. 1 and 2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4. Limit 3 to animals</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5. 3 not 4</td>
</tr>
<tr>
<td>EMBASE</td>
<td>1947 to week 27 of 2013</td>
<td>1. exp Panax/ or ginseng.mp.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2. exp Hemoglobin A, Glycosylated/ or exp Fructosamine/ or fructosamine.mp. or glycemia.mp or exp Hyperglycemia/ or fructosamine.mp. or diabetes mellitus.mp. or exp Insulin/ or dysoxemia.mp. or hyperinsulin.mp. or hyperinsulin*.mp. or exp Diabetes Mellitus, Type1/ or type 1 diabetes.mp. or exp Diabetes Mellitus, Type2/ or type 2 diabetes.mp. or glib albumin.mp. or exp Diabetes, Gestational/ or gestational diabetes.mp. or exp Prediabetic State/ or prediabetes.mp. or HBA1C.mp.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>3. 1 and 2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4. Limit 3 to animals</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5. 3 not 4</td>
</tr>
<tr>
<td>CINAHL</td>
<td>1985 to 3 July 2013</td>
<td>(Panax OR panax* OR ginseng* OR ninjin* OR renshen* OR shinseng* OR Ginsenosides OR Ginsenoids) AND (&quot;Glucose Tolerance Test&quot; OR OGTT OR Hemoglobin A, Glycosylated OR Hemoglobin A, Glycosylated OR HBA1C OR Fructosamine OR insulin* OR Glucose OR Hyperglycemia OR Hyperglycaemia OR glycaemia OR hyperinsulin* OR diabetes OR &quot;Diabetes Mellitus&quot; OR HOMA* OR glycemia OR &quot;gly* albumin&quot; OR diabetes OR &quot;metabolic syndrome&quot; OR &quot;homeostasis model assessment&quot; OR hyperglycemic OR hyperglycaemic)</td>
</tr>
<tr>
<td>The Cochrane Library</td>
<td>Through to 3 July 2013</td>
<td>(Panax OR panax* OR ginseng* OR ninjin* OR renshen* OR shinseng* OR Ginsenosides OR Ginsenoids) AND (&quot;Glucose Tolerance Test&quot; OR OGTT OR Hemoglobin A, Glycosylated OR Hemoglobin A, Glycosylated OR HBA1C OR Fructosamine OR insulin* OR Glucose OR Hyperglycemia OR Hyperglycaemia OR glycaemia OR hyperinsulin* OR diabetes OR &quot;Diabetes Mellitus&quot; OR HOMA* OR glycemia OR &quot;gly* albumin&quot; OR diabetes OR &quot;metabolic syndrome&quot; OR &quot;homeostasis model assessment&quot; OR hyperglycemic OR hyperglycaemic)</td>
</tr>
</tbody>
</table>

*The initial search for all databases was conducted on October 24 2012. All databases underwent updated searches on December 12 2012, March 5 2013, and July 3, 2013.
Table 2. Characteristics of studies investigating the effect of ginseng on glycemic outcomes

<table>
<thead>
<tr>
<th>Study *</th>
<th>Subject characteristics †</th>
<th>Age (y)</th>
<th>BMI (kg/m²) †</th>
<th>Setting</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Diabetes</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chio et al. 1997(^{238})</td>
<td>31 subjects with T1DM (15M:16F)</td>
<td>T:51.6 (10.9) C:50.4 (7.5)</td>
<td>N/R</td>
<td>OP, Korea</td>
</tr>
<tr>
<td>Kim et al. 2011(^{240})</td>
<td>38 subjects with T2DM (23M:15F)</td>
<td>T:56.0 (7.2) C:51.2 (10.7)</td>
<td>T:24.8 (2.7) C:23.9 (3.0)</td>
<td>OP, Korea</td>
</tr>
<tr>
<td>Ma et al. 2008(^{192})</td>
<td>20 subjects with T2DM (12M:8F)</td>
<td>51.0 (8.5)</td>
<td>28.5 (5.8)</td>
<td>OP, Hong Kong</td>
</tr>
<tr>
<td>Reeds et al. 2011 (^{193})</td>
<td>15 subjects with pre-diabetes and T2DM (1M:14F)</td>
<td>46.0 (11.6)</td>
<td>T1:35.0 (6.7) T2:36.0 (4.5) C:31.0 (2.2)</td>
<td>OP, USA</td>
</tr>
<tr>
<td>Sotaniemi et al. 1995(^{12})</td>
<td>36 subjects with T1DM (16M:20F)</td>
<td>T1:59.7 (7.0) T2:57 (9.0) C:60 (6.0)</td>
<td>N/R</td>
<td>OP, Finland</td>
</tr>
<tr>
<td>Vuksan et al. 2000(^{14})</td>
<td>24 subjects with T2DM (13M:11F)</td>
<td>64.0 (7.0)</td>
<td>28.0 (5.0)</td>
<td>OP, Canada</td>
</tr>
<tr>
<td>Vuksan et al. 2008(^{15})</td>
<td>19 subjects with T2DM (11M:8F)</td>
<td>64.0 (8.7)</td>
<td>28.9 (6.1)</td>
<td>OP, Canada</td>
</tr>
<tr>
<td>Yoon et al. 2012(^{194})</td>
<td>72 subjects with T2DM (44M:28F)</td>
<td>T1:52.7 (11.0) T2:52.7 (10.0) T3:51.1 (8.6) C:64 (10.0)</td>
<td>T1:26.3 (4.8) T2:24.0 (2.6) T3:25.4 (2.7) C:25.3 (1.9)</td>
<td>OP, Korea</td>
</tr>
<tr>
<td>Zhang et al. 2007(^{243})</td>
<td>84 subjects with pre-diabetes and T2DM (51M:33F)</td>
<td>T:62.9 (12.0) C:63.4 (10.5)</td>
<td>N/R</td>
<td>IP, China</td>
</tr>
<tr>
<td><strong>Non-diabetes</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dickman et al. 2009(^{239})</td>
<td>25 Otherwise healthy females</td>
<td>T:62.3 (5.9) C:61.7 (6.5)</td>
<td>T:25.3 (3.8) C:24.7 (2.5)</td>
<td>OP, USA</td>
</tr>
<tr>
<td>Park et al. 2012(^{241})</td>
<td>48 Females with pre-diabetes</td>
<td>T:43.1 (10.6) C:46.2 (11.0)</td>
<td>N/R</td>
<td>OP, Korea</td>
</tr>
<tr>
<td>Reay et al. [1] 2009(^{191})</td>
<td>23 Otherwise healthy subjects (12M:11F)</td>
<td>35.6 (1.1)</td>
<td>N/R</td>
<td>OP, UK</td>
</tr>
<tr>
<td>Reay et al. [2] 2009(^{191})</td>
<td>14 Otherwise healthy subjects (5M:9F)</td>
<td>38.4 (10.6)</td>
<td>N/R</td>
<td>OP, UK</td>
</tr>
<tr>
<td>Rhee et al. 2011(^{242})</td>
<td>64 subjects with EHPT (28M:36F)</td>
<td>T:55.0 (9.0) C:58.0 (6.0)</td>
<td>T:24.9 C:24.7</td>
<td>OP, Korea</td>
</tr>
<tr>
<td>Scaglione et al. 1996(^{243})</td>
<td>227 Otherwise healthy subjects (132M:95F)</td>
<td>T:48.0 (16.4) C:48.5 (16.5)</td>
<td>T:23.5 (1.2) C:23.4 (1.2)</td>
<td>OP, Italy</td>
</tr>
<tr>
<td>Shin et al. 2011(^{244})</td>
<td>30 subjects with Pre-diabetes (18M:12F)</td>
<td>T:47.1 (10.8) C:45.9 (10.5)</td>
<td>T:24.9 (7.4) C:22.6 (9.3)</td>
<td>OP, Korea</td>
</tr>
</tbody>
</table>
Table 2. Continued

<table>
<thead>
<tr>
<th>Glucose (mmol/L)</th>
<th>Insulin (pmol/L)</th>
<th>HbA1c (%)</th>
<th>HOMA-IR</th>
<th>Design</th>
<th>Ginseng form</th>
<th>Ginseng dose</th>
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</thead>
<tbody>
<tr>
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<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>T:10.9 (3.1)</td>
<td>T:9.0 (1.8)</td>
<td>T:9.0 (1.8)</td>
<td>Parallel</td>
<td>Capsule</td>
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<td>2.7g/d</td>
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<tr>
<td>C:13.2 (4.4)</td>
<td>C:10.1 (2.6)</td>
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<tr>
<td>T:7.6 (0.9)</td>
<td>T:6.59 (21.5)</td>
<td>T:7.4 (1.0)</td>
<td>Parallel</td>
<td>Capsule</td>
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<td>780.0mg/d</td>
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<tr>
<td>C:8.1 (1.5)</td>
<td>C:6.39 (20.8)</td>
<td>C:7.5 (1.2)</td>
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<tr>
<td>8.3 (3.1)</td>
<td>149 (111.9)</td>
<td>7.4 (3.8)</td>
<td>Crossover</td>
<td>Capsule</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>2214.0mg/d</td>
</tr>
<tr>
<td><strong>5.2 (0.2)</strong></td>
<td>52.4 (22.5)</td>
<td>5.9 (0.4)</td>
<td>Parallel</td>
<td>Capsule</td>
<td>T1: 3.0g/d</td>
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<tr>
<td></td>
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<td></td>
<td></td>
<td>first 2wks, 8.0g/d following 2wks.</td>
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</tr>
<tr>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>T2: 3.0g/d</td>
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</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>first 2wks, 8.0g/d following 2wks.</td>
<td></td>
</tr>
<tr>
<td><strong>8.3 (1.3)</strong></td>
<td>.</td>
<td>6.5 (1.7)</td>
<td>Parallel</td>
<td>Capsule</td>
<td>T1: 100.0mg/d</td>
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<td>T2: 200.0mg/d</td>
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</tr>
<tr>
<td><strong>8.3 (2.5)</strong></td>
<td>88.1 (66.9)</td>
<td>7.1 (0.1)</td>
<td>5.4 (3.9)</td>
<td>Crossover</td>
<td>Capsule</td>
<td>3.0g/d</td>
</tr>
<tr>
<td><strong>7.7 (1.7)</strong></td>
<td>35.0 (17.4)</td>
<td>6.5 (0.3)</td>
<td>1.7 (0.9)</td>
<td>Crossover</td>
<td>Capsule</td>
<td>6.0g/d</td>
</tr>
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<tr>
<td><strong>Non-diabetes</strong></td>
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<td><strong>4.6 (0.5)</strong></td>
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<td>Parallel</td>
<td>Capsule</td>
<td>1.0g/d</td>
</tr>
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<tr>
<td><strong>4.9 (0.9)</strong></td>
<td>72.9 (28.0)</td>
<td>5.5 (0.3)</td>
<td>.</td>
<td>Crossover</td>
<td>Capsule</td>
<td>200.0mg/d</td>
</tr>
<tr>
<td><strong>5.7 (0.5)</strong></td>
<td>97.2 (36.8)</td>
<td>5.5 (0.36)</td>
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<td>Crossover</td>
<td>Capsule</td>
<td>200.0mg/d</td>
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<tr>
<td><strong>5.3 (0.7)</strong></td>
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<td>Parallel</td>
<td>Capsule</td>
<td>200mg/d</td>
</tr>
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<tr>
<td><strong>5.7 (0.6)</strong></td>
<td>.</td>
<td>.</td>
<td>.</td>
<td>Parallel</td>
<td>Capsule</td>
<td>1.8g/d</td>
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</table>
### Table 2. Continued

<table>
<thead>
<tr>
<th>Ginseng preparation</th>
<th>Ginseng species</th>
<th>Comparator †</th>
<th>Follow-up</th>
<th>MQS **</th>
<th>Manufacturer</th>
<th>Funding Sources ††</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Diabetes</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Korean red ginseng (Unspecified preparation method)</td>
<td>Panax ginseng</td>
<td>No ginseng</td>
<td>24 wks</td>
<td>8</td>
<td>N/R</td>
<td>N/R</td>
</tr>
<tr>
<td>Fermented Korean red ginseng (Unspecified preparation method)</td>
<td>Panax ginseng</td>
<td>Placebo</td>
<td>12 wks</td>
<td>6</td>
<td>N/R</td>
<td>Agency</td>
</tr>
<tr>
<td>Roots of Panax ginseng</td>
<td>Panax ginseng</td>
<td>Placebo</td>
<td>4 wks</td>
<td>8</td>
<td>N/R</td>
<td>Agency</td>
</tr>
<tr>
<td>T1: Extract of Korean red ginseng T2: Ginsenoside Re</td>
<td>Panax ginseng</td>
<td>Placebo</td>
<td>4 wks</td>
<td>10</td>
<td>Spectrum laboratories, Gardena AIP0P, Gangdawon-Do, Korea</td>
<td>Agency</td>
</tr>
<tr>
<td>Extract of unspecified ginseng type</td>
<td>Unspecified</td>
<td>Placebo</td>
<td>8 wks</td>
<td>7</td>
<td>Dansk droge, Copenhagen, Denmark</td>
<td>N/R</td>
</tr>
<tr>
<td>Extract of American ginseng (CNT2000)</td>
<td>Panax quinquefolius</td>
<td>Placebo</td>
<td>8 wks</td>
<td>7</td>
<td>Chai-Na-Tai Corporation, Langley, BC, Canada</td>
<td>Industry</td>
</tr>
<tr>
<td>Rootlets of Korean red ginseng</td>
<td>Panax ginseng</td>
<td>Placebo</td>
<td>12 wks</td>
<td>8</td>
<td>Korea Ginseng Manufacturing Plant, Chung-buk, Korea</td>
<td>Agency</td>
</tr>
<tr>
<td>Vinegar extract of Panax ginseng (Ginsam)</td>
<td>Panax ginseng</td>
<td>Placebo</td>
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<td>10</td>
<td>YuYu Pharmaceuticaul Seoul, Korea</td>
<td>Industry</td>
</tr>
<tr>
<td>Extract of Panax quinquefolius saponin (PQS)</td>
<td>Panax quinquefolius</td>
<td>No Ginseng</td>
<td>4 wks</td>
<td>8</td>
<td>Manufacturer name reported in Chinese</td>
<td>Agency</td>
</tr>
<tr>
<td><strong>Non-diabetes</strong></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Dry whole root of American ginseng</td>
<td>Panax quinquefolius</td>
<td>Placebo</td>
<td>16 wks</td>
<td>7</td>
<td>Kaiser Farms, Wausau, Wi</td>
<td>Agency</td>
</tr>
<tr>
<td>Roots of Korean red ginseng</td>
<td>Panax ginseng</td>
<td>Placebo</td>
<td>12 wks</td>
<td>8</td>
<td>Korea Ginseng Corporation, Seoul, Korea</td>
<td>Agency</td>
</tr>
<tr>
<td>Extract of Panax ginseng (G115)</td>
<td>Panax ginseng</td>
<td>Placebo</td>
<td>~8wks</td>
<td>10</td>
<td>Pharmaton SA, Lugano, Switzerland</td>
<td>Industry</td>
</tr>
<tr>
<td>Extract of Panax ginseng (Cheong Kwan Jang)</td>
<td>Panax ginseng</td>
<td>Placebo</td>
<td>~8wks</td>
<td>10</td>
<td>Korea Ginseng Corporation, Seoul, Korea</td>
<td>Industry</td>
</tr>
<tr>
<td>Extract of Korean red ginseng</td>
<td>Panax ginseng</td>
<td>Placebo</td>
<td>12 wks</td>
<td>7</td>
<td>Korea Ginseng Co, Daejeon, Korea</td>
<td>Agency &amp; Industry</td>
</tr>
<tr>
<td>Extract of Panax ginseng (G115)</td>
<td>Panax ginseng</td>
<td>Placebo</td>
<td>12 wks</td>
<td>8</td>
<td>Pharmaton SA, Lugano, Switzerland</td>
<td>N/R</td>
</tr>
<tr>
<td>Extract of Korean red ginseng with cheonggukjang (fermented soybean)</td>
<td>Panax ginseng</td>
<td>Cheonggukjang (fermented soybean)</td>
<td>8 wks</td>
<td>10</td>
<td>Keimyung Foodex Co, Daegu, Korea</td>
<td>Agency</td>
</tr>
</tbody>
</table>
Table legend:

Abbreviations: EHPT = Essential hypertension; F = Female; M = Male; BMI = Body mass index; C = Control group; T = Treatment group; T1 = Treatment group #1; T2 = Treatment group #2; T3 = Treatment group #3; T1DM = Type 1 diabetes mellitus; T2DM = Type 2 diabetes mellitus; IP = Inpatient; OP = Outpatient; MQS = Heyland Methodological Quality Score; N/R = Not reported.

*Studies by Sotaniemi et al., Yoon et al., and Reeds et al. contained multiple comparisons, and to mitigate unit-of-analysis error, we combined groups to create a single pairwise comparison.
†Pre-DM included subjects with either Impaired Fasting Glucose or Impaired Glucose Tolerance.
‡Pre-study baseline BMI is listed. The study by Rhee et al. did not report SD for the mean BMI of participants.
§Pre-study baseline endpoints are listed. In studies were these values were not reported, the start value of control was assumed to be equivalent to baseline and was reported. Where start of control value was not given, end of control value was assumed to be equivalent to baseline and was reported. Assumed values are reported in bold. The study by Reay et al. [1] & [2] used n=23 and n=14 respectively for reporting data on fasting blood glucose, n=17 and n=12 respectively for reporting data on fasting plasma insulin, and n=18 and n=11 respectively for reporting data on HbA1c.
||Ginseng dose is reported individually for trials with multiple treatment groups.
¶All ginseng doses were compared to placebo, a control group that did not receive ginseng, or fermented soybean.
**Study quality was assessed by the Heyland Methodological Quality Score (MQS) and trials with a score ≥8 were considered to be of high quality.
††Agency funding is that from government, university or not-for-profit healthy agency sources. None of the trialists declared any conflicts of interest.
All data is expressed as mean (SD)
Table 3. Study quality assessment by the Heyland MQS*

<table>
<thead>
<tr>
<th>Study</th>
<th>Methods†</th>
<th>Sample‡</th>
<th>Intervention§</th>
<th>MGS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Randomization (n/2)</td>
<td>Blinding (n/1)</td>
<td>Analysis (n/2)</td>
<td>Selection (n/1)</td>
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<td>Diabetes</td>
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<td>Chio et al. 1997</td>
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<td>Ma et al. 2008</td>
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<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Reeds et al. 2011</td>
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<tr>
<td>Sakaniemi et al. 1995</td>
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<td>Vuksan et al. 2000</td>
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<td>Yoon et al. 2012</td>
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<td>Zhang et al. 2007</td>
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<td>Park et al. 2012</td>
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<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Reay et al. [1] 2009</td>
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<td>1</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Reay et al. [2] 2009</td>
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<td>1</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Rhee et al. 2011</td>
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</tr>
<tr>
<td>Scaglione et al. 1996</td>
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<td>0</td>
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</tr>
<tr>
<td>Shin et al. 2011</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>1</td>
</tr>
</tbody>
</table>

*The Heyland MQS assigns a score of 0 or 1 or from 0 to 2 over 9 categories of quality related to study design, sampling procedures, and interventions, for a total of 13 points. Trials that scored ≥8 were considered to be of higher quality236.

†Randomization was scored 2 points for being randomized with the methods described, 1 point for being randomized without the methods described, or 0 points for being neither randomized nor having the methods described. Blinding was scored 1 point for being double-blind or 0 points for “other.” Analysis was scored 2 points for being intention-to-treat; all other types of analyses scored 0 points.

‡Sample selection was scored 1 point for being consecutive eligible or 0 points for being preselected or indeterminate. Sample comparability was scored 1 point for being comparable or 0 points for not being comparable at baseline. Follow-up was scored 1 point for being 100% or 0 points for <100%.

§Treatment protocol was scored 1 point for being reproducibly described or 0 points for being poorly described. Co-interventions were scored 2 points for being described and equal, 1 point for being described but unequal or indeterminate, or 0 points for not being described. Treatment crossovers (where participants were switched from the control treatment to the experimental treatment) were scored 2 points for being ≥10%, 1 point for being <10%, and 0 points for not being described.
Table 4. Risk of bias assessment by the Cochrane risk of bias tool*

<table>
<thead>
<tr>
<th>Study</th>
<th>Sequence Generation †</th>
<th>Allocation Concealment ‡</th>
<th>Blinding §</th>
<th>Incomplete Outcome Data ǁ</th>
<th>Selective Outcome Reporting ¶</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diabetes</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Chio et al. 1997</td>
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<td>URB</td>
<td>HRB</td>
<td>LRB</td>
<td>URB</td>
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<td>Kim et al. 2011</td>
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<td>URB</td>
<td>LRB</td>
<td>HRB</td>
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<td>Vuksan et al. 2008</td>
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<td>Yoon et al. 2012</td>
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<td>LRB</td>
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<tr>
<td>Zhang et al. 2007</td>
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<td>HRB</td>
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<td>Non-diabetes</td>
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<td>URB</td>
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<tr>
<td>Park et al. 2012</td>
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<td>LRB</td>
<td>LRB</td>
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<td>Reay et al. [1] 2009</td>
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<td>Reay et al. [2] 2009</td>
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<td>Scaglione et al. 1996</td>
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<td>LRB</td>
<td>URB</td>
</tr>
</tbody>
</table>

Abbreviations: URB denotes unclear risk of bias; LRB low risk of bias; HRB high risk of bias.

*Studies were rated URB if insufficient information was given to assess risk; LRB if the study design is likely to have little influence over the true outcome; HRB if the study design is likely to have an influential effect on the true outcome.

†Sequence generation assessed whether the method of randomization was described.

‡Allocation concealment assessed whether investigators could tell which treatment participants were going to be randomized to.

§Blinding assessed whether the study was blinded to investigators and/or participants.

ǁIncomplete data assessed whether missing outcome data affected true outcome.

¶Selected outcome reporting assessed whether investigators pre-registered trial and/or specified primary and secondary outcomes.
Table 5. Continuous meta-regression analysis for the effect of ginseng on glycemic parameters

<table>
<thead>
<tr>
<th>Subgroup</th>
<th>Range</th>
<th>No. of trials</th>
<th>N</th>
<th>$\beta$ [95% CI]</th>
<th>Residual $I^2$</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>FBG</strong> Baseline (mmol/L)</td>
<td>4.5 - 13.2</td>
<td>16</td>
<td>770</td>
<td>-0.26 [-0.40, -0.13]</td>
<td>70.4%</td>
<td>0.001*</td>
</tr>
<tr>
<td>Duration (weeks)</td>
<td>4 - 24</td>
<td>16</td>
<td>770</td>
<td>-0.02 [-0.11, 0.07]</td>
<td>89.7%</td>
<td>0.66</td>
</tr>
<tr>
<td><strong>FPI</strong> Baseline (pmol/L)</td>
<td>41 - 149</td>
<td>10</td>
<td>347</td>
<td>0.03 [-0.33, 0.40]</td>
<td>29.3%</td>
<td>0.85</td>
</tr>
<tr>
<td>Duration (weeks)</td>
<td>4 - 12</td>
<td>10</td>
<td>347</td>
<td>-1.19 [-2.80, 0.42]</td>
<td>7.1%</td>
<td>0.13</td>
</tr>
<tr>
<td><strong>HbA1c</strong> Baseline (%)</td>
<td>5.4 - 9.6</td>
<td>9</td>
<td>264</td>
<td>-0.11 [-0.28, 0.06]</td>
<td>51.7%</td>
<td>0.18</td>
</tr>
<tr>
<td>Duration (weeks)</td>
<td>4 - 24</td>
<td>9</td>
<td>264</td>
<td>-0.009 [-0.05, 0.03]</td>
<td>52.3%</td>
<td>0.61</td>
</tr>
<tr>
<td><strong>HOMA-IR</strong> Baseline</td>
<td>1.04 -7.4</td>
<td>7</td>
<td>303</td>
<td>-0.25 [-0.73, 0.23]</td>
<td>83.6%</td>
<td>0.24</td>
</tr>
<tr>
<td>Duration (weeks)</td>
<td>4 - 12</td>
<td>7</td>
<td>303</td>
<td>0.13 [-0.14, 0.40]</td>
<td>81.5%</td>
<td>0.28</td>
</tr>
</tbody>
</table>

Abbreviations: FBG: Fasting blood glucose; FPI: Fasting plasma insulin; HbA1c: Glycated hemoglobin; HOMA-IR: Homeostasis model assessment of insulin resistance.

Changes in mean-difference between ginseng and control intervention per unit change in each predictor using continuous meta-regressions. A positive $\beta$-coefficient implies an increase in FBG, FPI, HbA1c, or HOMA-IR response with ginseng supplementation relative to control; and a negative $\beta$-coefficient implies a decrease in FBG, FPI, HbA1c, or HOMA-IR response with ginseng supplementation relative to control.

*Indicates significance at $P < 0.05$. 
CHAPTER 5

ACUTE DOSE RESPONSE EFFECTS OF KOREAN WHITE GINSENG (PANAX GINSENG C.A. MEYER) ON GLYCEMIC AND VASCULAR PARAMETERS IN INDIVIDUALS WITH TYPE 2 DIABETES: AN ACUTE, RANDOMIZED, DOUBLE BLIND, PLACEBO-CONTROLLED, MULTIPLE-CROSSOVER TRIAL.
5. ACUTE DOSE RESPONSE EFFECTS OF KOREAN WHITE GINSENG (PANAX GINSENG C.A. MEYER) ON GLYCEMIC AND VASCULAR PARAMETERS IN INDIVIDUALS WITH TYPE 2 DIABETES: AN ACUTE, RANDOMIZED, DOUBLE BLIND, PLACEBO-CONTROLLED, MULTIPLE-CROSSOVER TRIAL.

Running title: Efficacy of Korean white ginseng on vascular and glycemic health

Esra’ Shishtar¹ ², Elena Jovanovski¹ ², Alexandra Jenkins¹ ², Vladimir Vuksan†¹ ²

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²Department of Nutritional Sciences, Faculty of Medicine, University of Toronto, Toronto, ON

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Funding source:

This research was supported by a grant from the Department of Herbal Crop Research; National Institute of Horticultural and Herbal Science, RDA, Korea

Manuscript status: Submitted to Clinical Nutrition Research
5.1 ABSTRACT

Background: Korean red ginseng (steam treated variety of \( Panax ginseng \) C.A. Meyer), one of the most widely used herbal remedies, has been clinically shown to improve certain CVD risks when paired with orthodox medicine. Whether this holds true for the dried non-steamed variety, known as Korean white ginseng (KWG), is unclear. Therefore, this exploratory study investigated the efficacy and safety of increasing doses of KWG on glycemic and vascular parameters in individuals with type 2 diabetes mellitus (T2DM).

Methods: Using an acute, randomized, placebo-controlled, double-blind, crossover design, 25 participants with well-controlled T2DM (12-males:13-females, age:63±9 years, A1c:6.9±0.7%, BMI:29.3±4.3kg/m\(^2\)) underwent five visits during which they received 1g, 3g, or 6g KWG or 3g wheat bran control (twice) together with 50g-glucose load. The protocol followed the OGTT guidelines with capillary blood samples drawn at 0, 15, 30, 45, 60, 90, 120, and 180min after ginseng administration. For the duration of 240minutes, Al, and central blood pressure (CBP) were measured at baseline and at 60min-intervals, and ambulatory blood pressure (ABP) was assessed at baseline and at 10min-intervals. A symptoms questionnaire was used to assess safety and adverse events.

Results: Two-way ANOVA demonstrated a significant time-treatment interaction effect on Al \((P=0.01)\) with one-way ANOVA showing significant reductions in Al with 3g KWG relative to control \((P=0.04)\). Compared to control, acute administration of KWG appeared to be safe, but did not affect any other postprandial glycemic or vascular parameters.

Conclusions: KWG might have a beneficial effect on Al, a cumulative indicator of arterial health. However, these results are preliminary and highlight the need for long-term investigation with a focus on its potential accountable components.

Clinical Trial Registration: NCT01699074
Keywords: Clinical trial, Korean white ginseng, type 2 diabetes, postprandial blood glucose, central blood pressure, brachial blood pressure, augmentation index
5.2 INTRODUCTION

Korean ginseng, the fleshy roots of *Panax ginseng* C.A. Meyer, is a highly valued medicinal herb in the orient with a history of use that dates back to more than 3000 years. There is growing evidence in the literature on the potential benefits of *Panax ginseng* C.A. Meyer—the most commonly consumed, and widely researched ginseng species—on glycemic and vascular health. Based upon the processing it undergoes, Korean ginseng exists in two forms: white ginseng, a 4 - 6 year root that is dried after peeling, and red ginseng, a 6 year root that is steam treated prior to drying. Steaming the fresh ginseng roots has been traditionally performed for the purposes of preserving and extending its shelf life. Although findings indicate that steaming increases ginseng’s pharmacological potency, it has been also shown that high temperature associated with steaming significantly alters its ginsenosides, as well as results in the loss of malonyl type ginsenosides. Given that much of ginseng’s therapeutic properties are attributed to its ginsenosides, it is very likely that the steaming application may lead to the loss of its biological properties, and hence, its medicinal benefits.

Favorable metabolic actions of KWG, including anti-obesity and anti-diabetic effects, have been reported in animal models. However, such effects have not yet been investigated in the human population. In light of the growing worldwide epidemic of diabetes and its associated CVD complications, combined with frequent failure of meeting treatment goals, there is a compelling argument for alternative prevention and treatment options to address multifaceted factors in CVD prevention. Provided that KWG has shown ameliorating effects on certain cardiometabolic functions pre-clinically, combined with the high-steaming costs of KRG, KWG might present as a promising therapeutic candidate for improving certain CVD risks, and thus offer a less-expensive natural health alternative relative to KRG. The present study,
therefore, aims to assess the effect of KWG in a dose-dependent manner on glycemic and vascular parameters in subjects with T2DM through a randomized, double-blind, placebo-controlled, multiple-crossover design. KWG will be investigated at doses 1, 3, and 6g, allowing for determination of a potential optimal dose for possible long term exploration.

5.3 METHODS

5.3.1 Participants

Thirty participants with T2DM were recruited through newspaper advertisement and research database. All gave informed written consent to take part in the study (Appendix 1), which was approved by the Research Ethics Board at St Michael’s Hospital (Appendix 2). Inclusion criteria included individuals with presence of T2DM (A1c 6.5-8.5%) for at least 1 year, treated with diet alone or diet and oral hypoglycemic medication that was unchanged starting at least three months prior to the study. In addition, eligible participants were between the ages 18-75 years, body mass index (BMI) between 25-35 kg/m², systolic blood pressure (SBP) <160 mmHg and diastolic blood pressure (DBP) <100 mmHg, clinically euthyroid, normal renal and hepatic functions; no major illness, non-pregnant, not taking herbs or supplements, no excessive alcohol (>3 drinks/day) or cigarette use (>10 cigarettes/day), no allergy or sensitivity to the study interventions or gelatin used in the capsules. The investigational products were approved by the NHPs Directorate of Health Canada (Appendix 3). All study dosing visits were carried out at the Risk Factor Modification Centre, St. Michael’s Hospital (Toronto, Canada).

5.3.2 Treatments

The four treatment interventions included 1g, 3g, and 6g of 4-year old dried ginseng root powder, and 3g wheat bran control. All doses of KWG were provided by the Department of Herbal Crop Research; National Institute of Horticultural & Herbal Science, RDA, Korea, in a set of twelve 500mg opaque #00 white gelatin capsules. All study materials were tested for
microbial contamination, heavy metal and pesticides, and fell within the predetermined acceptable ranges set by Health Canada\textsuperscript{255}. The wheat bran control was purchased from the American Association of Cereal Chemists International (St. Paul, Minnesota, USA), was ground into dried powder, and encapsulated into 500mg opaque #00 white gelatin capsules. All capsules were identical in appearance. The weighing and encapsulation of the wheat bran control, as well as the blinding of all treatments were performed by an individual otherwise not involved in the study in order to ensure the blinding of study personnel and subjects. Shelf-life of capsules was approximately 2 years. Randomization to treatment was done using a computer-generated random number table. Subjects were assigned to consecutive numbers after they provided written informed consent.

5.3.3 Study Protocol

Each subject underwent five separate study visits in a randomized order after a 10-12 hour overnight fast, at the Risk Factor Modification Centre at St. Michael’s Hospital (wheat bran, the study control, was administered twice). Each visit was at least 4 days apart to allow for treatment washout between visits. Prior to the study period, subjects were asked to maintain a constant level of physical activity and their usual diet during the course of the study. Compliance with these conditions was assessed at each visit using a pre-clinical information questionnaire (Appendix 4). At the start of each study visit, the following measurements were taken on a clinical assessment form (Appendix 5): height (only at first visit), weight (TANITA BC-418 Segmental Body Composition Analyzer), waist circumference, and body fat percentage (TANITA BC-418 Segmental Body Composition Analyzer). Subjects were then taken to the exam room, where they filled out a pre-clinical information questionnaire and remained seated for at least 10 minutes to achieve resting heart rate and BP, after which 3 consecutive baseline systolic and diastolic brachial BP readings (OMRON Digital Automatic Blood Pressure Monitor HEM-907) were
obtained. Next, CBP and Al were measured using SphygmoCor Vx instrument (Atcor Medical, Australia). After that, subjects were fitted with an ambulatory blood pressure monitor (ABPM), which was initialized to obtain automatic blood pressure readings every 10 minutes post-treatment for duration of 240 minutes. Following vascular measures, a capillary baseline blood sample was obtained. After these baseline measurements, subjects consumed orally, in a random, double blind fashion, 12 capsules containing either wheat bran or KWG along with a 50-g oral glucose challenge. Finger prick blood samples were subsequently taken at 15, 30, 45, 60, 90, 120 and 180 min post-treatment and analyzed for plasma glucose levels. AI and CBP were measured at 60, 120,180 and 240 minutes post-treatment. After the third hour post-treatment, subjects consumed a standardized breakfast, which included 200 mL water, 15 g (1 tbsp) low fat cream cheese (Philadelphia, Kraft Canada Inc), and 1 slice whole wheat toast (Dempster’s 100% whole wheat bread). For the duration of the visit, occurrence of adverse events was documented on a validated symptoms questionnaire in which 100 mm visual analog scale was used (Appendix 6). At the end of each visit, a 24-hour symptoms form was given which was returned at the next scheduled visit (Appendix 7). One day after the last (fifth) visit, the study coordinator contacted all subjects to ensure they report any adverse symptoms (Appendix 8).

### 5.3.4 Ginsenoside analyses

The ginsenoside profile of the 4 year-old KWG root was analyzed by the Department of Herbal Crop Research; National Institute of Horticultural & Herbal Science; RDA; Korea, using thin-layer chromatography technique. The total ginsenoside concentrations in the 4yr-old KWG root was 1.8%. The individual concentrations for the PPD and PPT ginsenosides, along with their relevant ratios are shown in Table 1.
Table 1. Ginsenoside profile of the study investigational material, Korean white ginseng root. Results are presented as mean of triplicate analysis

<table>
<thead>
<tr>
<th>Ginsenosides</th>
<th>Protopanaxadiols (PPD) (%wt/wt)</th>
<th>Protopanaxatriols (PPT) (%wt/wt)</th>
<th>Total (%wt/wt)</th>
<th>Ratios (%wt/wt: %wt/wt)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Rb\textsubscript{1} 0.236</td>
<td>Re 0.304</td>
<td>1.841</td>
<td>PPD:PPT 0.352</td>
</tr>
<tr>
<td></td>
<td>Rb\textsubscript{2} 0.073</td>
<td>Re 0.913</td>
<td></td>
<td>Rb\textsubscript{1}:Rg\textsubscript{1} 0.776</td>
</tr>
<tr>
<td></td>
<td>Rb\textsubscript{3} 0.006</td>
<td>Rf 0.142</td>
<td></td>
<td>Rb\textsubscript{2}:Rc 0.562</td>
</tr>
<tr>
<td></td>
<td>Rc 0.130</td>
<td>Rg\textsubscript{2} 0.002</td>
<td></td>
<td>Rg\textsubscript{1}:Re 0.333</td>
</tr>
</tbody>
</table>

5.3.5 Study measurements

5.3.5.1 Anthropometric Assessments

At every visit, subjects underwent anthropometric measurements including height (only at first visit), weight, waist circumference, and assessment of body composition. Height was measured with a wall-mounted stadiometer (Perspective Enterprises, Portage, MI) with head in the “Frankfurt horizontal” position in barefooted subjects and rounded to the nearest centimeter. Waist circumference was measured using a non-stretchable measuring tape positioned at the level of noticeable waist narrowing and recorded to the nearest centimeter. After emptying of bladder, removing any excess clothing and shoes, weight, and body composition (BMI, % body fat) were analyzed via Bioelectrical Impedance Analysis using the TANITA BC-418 Segmental Body Composition Analyzer (Arlington Heights, Illinois, USA). All of these measurements were recorded on a clinical assessment form (Appendix 5).

5.3.5.2 Oral Glucose Tolerance Test

A finger prick capillary blood sample was taken at baseline (time 0) after which a 300mL orange flavored 50g glucose drink (GLUCODEX® Ratiopharm Inc, Mississauga, Canada) was administered with 12 capsules of either wheat bran or 1,3,or 6g KWG over a period of five minutes. Additional blood samples were drawn at 15, 30, 45, 60, 90, 120 and 180 minutes after the start of the test drink. Whole blood samples (25-75μL) were collected in 7mL fluoride oxalate-treated flat base polystyrene tubes (Sarstedt Inc, Montreal, Canada) and immediately stored at -20°C pending analysis. Tubes were prepared with 375μg sodium fluoride and 300μg potassium oxalate and allowed to air dry for three days prior to use. Whole blood samples were analyzed for glucose concentration using the YSI 2300D STAT Plus Glucose & Lactate Analyzer.
(Yellow Springs, Ohio, USA) within three days of collection. The analytical method employed by the YSI is based on the oxidation of glucose leading to electron production and the subsequent generation of an electric current that is linearly proportional to the glucose concentration\textsuperscript{256}. The YSI was calibrated with a standard 10mmol/L glucose solution prior to and during analysis of each set of seven samples.

5.3.5.3 Baseline brachial blood pressure

Baseline brachial BP (BBP) was assessed oscillometrically using the OMRON Digital Automatic Blood Pressure Monitor HEM-907 (Bannockburn, Illinois, USA). Subjects remained seated in a quiet, temperature-controlled room for 5-10 minutes with arm supported at heart level to achieve resting heart rate and blood pressure. Subsequently, three readings were obtained from the brachial artery in the left forearm, with one minute separating each measurement. The arithmetic mean of the three readings was used in all baseline demographic BP data.

5.3.5.4 Arterial stiffness and central blood pressure

Subjects underwent a non-invasive measurement of arterial stiffness using applanation tonometry (SphygmoCor, AtCor Medical, West Ryde, Australia) at baseline, and 1, 2, 3, and 4 hour post treatment, where they were required to lie down in a supine position to minimize movement. A tonometer was used as a pressure sensor at the radial artery to obtain a measure of AI and CBP via pulse wave analysis using the software’s validated algorithm. As AI is closely dependent on heart rate, and in order to eliminate variation in heart rate when comparing acute effects of treatments, an index normalized for heart rate of 75 beats per minutes was used in accordance with Wilkinson et al\textsuperscript{257}. 
5.3.5.5 Brachial blood pressure via ambulatory blood pressure

ABP was monitored oscillometrically using the Spacelabs 90207 system (Mississauga, Ontario, Canada). The cuff was secured on the nondominant arm and measurements were taken at baseline, and at 10 minutes intervals post-treatment for the entire study visit (240 min), totaling 24 readings. The 24 BP measures were further meaned into a set of 4 readings, each representing hourly brachial BP readings (1, 2, 3, & 4hr), where each hourly BP reading was derived from an average of six 10-minute interval time points. Measurements were automatically repeated after 2 minutes if an error occurred or if obtained readings fell outside predefined acceptable ranges.

5.3.5.6 Questionnaires

A medical information form was administered at the screening visit. This form collected data on medical history, current medication use and daily exercise/diet regimens (Appendix 9). Compliance with the study protocol was assessed at every visit via a pre-clinical information questionnaire (Appendix 4). Because of the few references in the literature reporting on possible adverse effects of ginseng, including headache, insomnia, and nervousness, a symptoms questionnaire including these symptoms as well as other symptoms of daily discomfort was administered at every visit (Appendix 6). As well, at the end of each visit, a 24 hour symptoms form was given to the subjects to take and complete in the 24 hrs after their visit. They were required to bring the completed form on their next scheduled visit (Appendix 7). All subjects were contacted by the study coordinator after their last (5th) visit, should they develop any adverse symptoms (Appendix 8).
5.3.5.7 Study outcomes and statistical analysis

The primary outcome was the difference in the effect of KWG on PPBG iAUC relative to control. PPBG iAUC was calculated geometrically ignoring areas below the 0 value for each subject and were averaged for each intervention. The secondary outcomes included the mean changes of the following parameters relative to control: PPBG, AI, central SBP, central DBP, brachial SBP, and brachial DBP. Changes in PPBG measures were calculated at each capillary blood sampling time point (15, 30, 45, 60, 90, 120, and 180 minutes). Change in AI, normalized for a heart rate of 75 beats/minute, was calculated at 60, 120, and 180, and 240 minutes. Similarly, change in CBP was calculated at each hour time point across the 4 hr visit duration. The 24 brachial BP measures obtained at 10 minute intervals from 10 to 240 minutes inclusive were meaned into a set of 4 measures, each measure representing hourly brachial BP (1, 2, 3, & 4hr) readings, where every hr represents an average of six 10-minute interval time-points. Change in ABP was calculated at these hourly time-points. Statistical analyses were performed using the Statistical Package for the Social Sciences (SPSS) release 21.0 (SPSS Inc., Chicago, IL). All data was tested for normality. For normally distributed data, two-way repeated measures GLM ANOVA assessed the independent and interactive effects of treatments and protocol time on change in AI, CBP, ABP, and PPBG. All study outcomes underwent one-way repeated measures GLM ANOVA along with Tukey’s post hoc test, where differences between treatment-associated means at each individual time point were explored. Paired student t-test assessed differences in subjective ratings of symptoms for both the clinical testing and washout periods. Subject characteristics were expressed as mean ± standard deviation (SD), while all other data was presented as mean ± standard error of the mean (SEM). All data was considered statistically significant at P<0.05.
5.3.5.8 Sample size

Given that the study employs a crossover design, and based on our previous observations with similar study designs, a treatment difference of 130 min.mmol/L in PPBG iAUC (SD=150 min.mmol/L) was used for determination of sample size, indicating that approximately 23 individuals would be required (2 sided $\alpha=0.05$, 0.8 power). Assuming a 30% attrition rate, a total of 30 subjects were to be recruited.

5.4 RESULTS

5.4.1 Subject characteristics, compliance, and symptoms

A total of 30 subjects with T2DM provided written informed consent and were enrolled in the study, of which five dropped out due to time conflicts. Twenty-five subjects completed the study, 12 males and 13 females, mean±SD age: 63±9 years, BMI: 29.3±4.3kg/m², waist circumference: 101.7 ± 10.5 cm, fasting blood glucose: 7.4±0.05 mmol/L, A1C: 6.9±0.7%, resting SBP/DBP: 133±15.9/74 ± 7.5 mmHg. The mean duration of diabetes was 9±7 years while the onset of hypertension was 14±12 years, with 15 subjects taking glucose lowering and 9 subjects taking anti-hypertensive medications. All subjects consumed a minimum of 150g of carbohydrate over the 3 days prior to each test visit and refrained from consuming their medications on the morning of the study visits. None of the study subjects reported use of NHPs within three weeks prior to the screening visit, and no pregnancy cases were reported by any of the female participants both before and during the time of the study. None of the participants had any weight changes that exceeded 5% of their baseline measured weight. Additionally, all treatments were consumed by each participant based upon the initial treatment sequence that they were randomly assigned to, having at least 4 days washout between visits. The study
protocol was followed without difficulty, and participants reported no change in their overall lifestyle pattern including diet and physical activity throughout the duration of the study. There were also no differences in reported symptoms of bloating, belching, nausea, dizziness, headache, diarrhea, flatulence, polyuria, insomnia, anxiety, numbness, light-headedness, thirst, or cramping between KWG treatments and control during the clinic visits or the intervening washout period.

5.4.2 Effects of KWG treatments on glycemia

5.4.2.1 Postprandial blood glucose iAUC and postprandial blood glucose

Repeated measures one-way ANOVA did not demonstrate a significant effect in PPBG iAUC of KWG doses relative to control \((P=0.78)\) (Figure 1). As well, two-way repeated measures GLM ANOVA did not show a significant time-treatment interactive effect on PPBG \((P=0.38)\) (Figure 2), and repeated measures GLM one-way ANOVA further demonstrated that there were no significant differences in the PPBG responses between the three KWG doses and control at any individual time point.
Figure 1. The effect of KWG treatments compared to control on postprandial blood glucose iAUC

Blood glucose iAUC after 3 hours of intervention in 25 subjects. Control represents the mean response to two placebos. Values are mean ± SEM. PPBG iAUC, postprandial blood glucose incremental area under the curve; KWG, Korean white ginseng. One-way ANOVA: (P=0.78)
5.4.3 Effects of KWG on vascular parameters

5.4.3.1 Brachial blood pressure

Figure 3 and Figure 4 display the mean changes in BBP relative to baseline between the three KWG doses and control taken across 240 minutes post-treatment at 10 min intervals via an ABPM, after which they were averaged into a set of 4 hourly time points. Two-way repeated measures GLM ANOVA did not demonstrate a significant time-treatment interactive effect on brachial SBP (P=0.11) or brachial DBP (P=0.14) for the entire 240 minutes. Moreover, no significant differences occurred between the three KWG doses and control at any hour time point. The mean absolute brachial systolic and diastolic BP did not differ significantly between treatments at baseline and the data showed normal distribution.
Figure 3. The effect of KWG treatments versus control on brachial systolic blood pressure

Change from baseline in brachial systolic blood pressure across 4 hours of intervention in 25 subjects with T2DM. Control represents the mean response to two placebos. Values are mean ± SEM. BP, Blood pressure; KWG, Korean white ginseng. Two-way ANOVA (P=0.11)

Figure 4. The effect of KWG treatments versus control on brachial diastolic blood pressure

Change from baseline in brachial diastolic blood pressure across 4 hours of intervention in 25 subjects with T2DM. Control represents the mean response to two placebos. Values are mean ± SEM. BP, Blood pressure; KWG, Korean white ginseng. Two-way ANOVA (P=0.14)
5.4.3.2 Augmentation index and central blood pressure

Figure 5 shows the mean changes in AI relative to baseline between the three KWG doses and control taken at 60 minute post-treatment intervals from 0 to 240 minutes. Two-way repeated measures GLM ANOVA demonstrated a significant time-treatment interactive effect on AI, such that the effect of treatment was dependant on protocol time ($P=0.01$). Additionally, the effect of treatment was explored at individual time points of the AI curves using repeated measures GLM one-way ANOVA. The Tukey’s post hoc test showed that 3g KWG elicited significant reduction in AI at 240 minutes relative to control ($P=0.035$). The test also revealed that both 3g and 1g KWG were significantly different from the 6g KWG dose ($P=0.005$; $P=0.02$ respectively) at 240 minutes.

Figures 6 and 7 show the mean changes in central BP relative to baseline between the three KWG doses and control taken at 60 minute post-treatment intervals from 0 to 240 minutes. Two-way repeated measures GLM ANOVA did not demonstrate a significant time-treatment interactive effect on change in central SBP ($P=0.95$) or central DBP ($P=0.92$) for the entire 240 minutes. There were also no significant differences amongst the three KWG doses and control at each of the 60 minute intervals. The mean absolute AI, central SBP, and central DBP did not differ significantly between any of the treatments at baseline and the data showed normal distribution.
Figure 5. The effect of KWG treatments relative to control on augmentation index

Change from baseline in AI (n=25). Control represents the mean response to two placebos. Values are mean ± SEM. AI, Augmentation index; KWG, Korean white ginseng. Two-way ANOVA: Significant time x treatment interaction (P=0.01) *One-way ANOVA: Significant treatment effect between control and 3g KWG (P=0.04) ** One-way ANOVA: Significant treatment effect between 3g KWG and 6g KWG (P=0.005) *** One-way ANOVA: Significant treatment effect between 1g KWG and 6g KWG (P=0.02)
Figure 6. The effect of KWG treatments versus control on central systolic blood pressure

Change from baseline in central systolic blood pressure across 4 hours of intervention in 25 subjects with T2DM. Control represents the mean response to two placebos. Values are mean ± SEM. BP, Blood pressure; KWG, Korean white ginseng. Two-way ANOVA (P=0.95)

Figure 7. The effect of KWG treatments versus control on central diastolic blood pressure

Change from baseline in central diastolic blood pressure across 4 hours of intervention in 25 subjects with T2DM. Control represents the mean response to two placebos. Values are mean ± SEM. BP, Blood pressure; KWG, Korean white ginseng. Two-way ANOVA (P=0.92)
5.5 Discussion

We present here, for the first time, that KWG - the most commonly consumed form of ginseng in Korea\textsuperscript{23} - exhibits a favorable acute effect on AI, a marker of arterial stiffness, in individuals with T2DM. More specifically, we show that this effect is associated with the consumption of 3g KWG dose, and the effect appeared not be dose dependent. These findings are of interest given the well-established predictive ability of AI in determining future CVD events\textsuperscript{84,85}. Our results also indicate that KWG does not significantly reduce postprandial glycemia relative to control. A U-shaped relationship was observed between escalating doses of KWG and PPBG, with 3g KWG showing the greatest reduction, and 6g KWG eliciting an increase, albeit not significant.

An extensive number of clinical, laboratory and animal studies have investigated the therapeutic potential of ginseng in improving glycemia, with much focus being placed on two species: Asian, typically the steamed variety of Korean ginseng, and AG\textsuperscript{108,259}. Presently, a number of reports from animal models reveal a possible anti-diabetic potential of the non-steamed variety of Asian ginseng\textsuperscript{28,260-262}. Yet, to date, no randomized controlled trials have explored the acute glycemic and vascular effects of such varieties in T2DM. With respect to findings seen with other ginseng species and types, our results do not demonstrate a favorable acute glycemic effect as observed with the original efficacious batch of AG following standard OGTT in both healthy and subjects with T2DM\textsuperscript{181}. As well, our findings are inconsistent with the favorable postprandial glycemic results of Panax ginseng extracts (Ginsana G115) and KRG rootlets, seen either alone or with standard glucose tolerance tests in healthy individuals\textsuperscript{186,190}. On the other hand, we are only aware of two studies that have assessed the acute glycemic effects of the non-steamed Asian ginseng variety in healthy subjects. In these studies, both null and opposing glycemic effects were observed with single and combined ginseng doses following a 75g-OGTT\textsuperscript{30}. In relation to these two studies, we show similar null but not increasing glycemic
effects following acute administration of escalating KWG doses. Comparing the total ginsenoside profile of the present ginseng to the one used in these two studies reveals that the concentration in the present sample is nearly twice as much (1.8% vs. 0.8% ginsenosides). However, despite having greater ginsenoside levels, as well as including subjects with T2DM, no glycemic lowering benefits were observed. On the other hand, our neutral glycemic observations are in contrast with evidence from animal studies\textsuperscript{28,260}. In a recent 3 week investigation, it was found that the Malonyl ginsenosides, a type of ginsenosides that exist in both fresh and air dried (non-steamed) root, significantly lowered fasting blood glucose when given at 50 and 100 mg/kg/wt doses to high fat diet fed and streptozotocin-induced diabetic rats\textsuperscript{175}. As well, a previous study showed that 4-week oral administration of both white ginseng radix and rootlet lowered fasting blood glucose levels in KKAy mice compared to controls\textsuperscript{28}.

Reasons for the discrepancies in our glycemic findings are unclear. Variability in the ginsenoside profile might partially provide some explanation\textsuperscript{263}. Analysis of ginsenoside profile revealed that the total ginsenoside content (1.8%) of the current ginseng variety was almost half of that found in our original efficacious batch of AG (3.2%)\textsuperscript{181}, and the optimal PPD:PPT ratio was lower by 8-folds (0.35 vs. 2.44). Additionally, the concentration of individual PPD ginsenosides, including Rb1 and Rc, that have previously shown a glycemia lowering potential in animal models when provided at levels greater than 1, was not satisfied\textsuperscript{264}. The implication is that it is possible that the main ginsenosides that have previously demonstrated hypoglycemic effects were not meeting their efficacy range. In line with this paradigm, we can speculate that the observed U-shaped dose-response relationship may be allocated to the level of available bioactive components, representing the fundamental nature of a pharmacological agent where the biological activity is either below the threshold, as observed with 1g KWG, or above the threshold with the upper dose of 6g KWG. Taken together and though not significant from control, 3g KWG appears to be the most optimal dose of KWG tested that can potentially
improve glucose metabolism as it exhibited the most pronounced PPBG lowering effect amongst the investigated doses, which is in line with the dose range suggested within ginseng monographs based on traditional recommendations\textsuperscript{265}.

As it is well established that elevated baseline glycemia would generate greater reductions with anti-hyperglycemic agents\textsuperscript{61}, and given that a strong correlation exists between HbA1c and PPBG when HbA1c levels are relatively close to the optimal target value of $\leq 7\%$\textsuperscript{266-268}, the lack of improvements in glycemia with the presumably optimal dose, 3g KWG, could be possibly explained by the well controlled glycemic status of our study participants (HbA1c=6.9%), likely resulting from their underlying anti-diabetes medications. This in turn might have underestimated the hypoglycemic potential of KWG. Hence, the potential for KWG to improve glycemia should not be totally dismissed. Future trials exploring the anti-hyperglycemic effects of KWG in subjects with compromised metabolic function are therefore warranted.

Arterial stiffness has emerged as a novel marker of CVD risk, displaying a clear independent predictive value\textsuperscript{85,269}. As such, acute improvements in AI demonstrated with 3g KWG may have clinical significance, and could aid in broadening our understanding on the hemodynamic potential of ginseng.

The ameliorative acute effects seen on AI add to those from previous clinical trials though different ginseng species and varieties were explored in varying study populations. In an acute RCT, 3g KRG significantly lowered arterial stiffness in healthy individuals, relative to control, with no improvements in BP\textsuperscript{21}. Another recent study conducted in individuals with T2DM and concomitant hypertension showed that 3g AG significantly lowered radial AI and attenuated systolic BP after 12 weeks of consumption\textsuperscript{220}. Conversely, our results are not in line with data from two studies where 3g and 4.5g of KRG administered daily for 3 months did not show improvements on arterial stiffness in subjects with metabolic syndrome and hypertension \textsuperscript{195,216}. The inconsistency in the findings could be possibly attributed to the differences in the method by
which arterial stiffness was assessed in the current (pulse wave analysis) versus the two studies (brachial-ankle PWV).

The dose response patterns observed with our Al results at the 4hr time point, as similarly depicted in our postprandial glycemic findings, further illustrate the concept of a range for bioactivity and potential anti-complementary effects of ginsenosides outside of the efficacious range. We observed significant reductions with 3g KWG relative to control, in addition to detecting significant differences among both the 1g KWG and 3g KWG compared to 6g KWG, indicating that 6g KWG and 3g KWG possess the greatest potential to increase and decrease Al levels respectively. Consistent with our glycemic results, and despite not being significantly different from control, it appears that the level of ginsenosides in the 6g KWG dose may have exceeded the therapeutically viable threshold rendering opposing effects. Similarly, although the 1g KWG dose did show modest reductions on Al, being significantly different from the 6g KWG dose, it seems that it is not delivering sufficient amount of bioactives, or ginsenosides, in order to elicit a pronounced effect. As a parallel to our glycemic results and though not showing significant reductions on postprandial glycemia, the 3g KWG dose did indeed demonstrate significant improvements on Al, relative to control. Taken together, we can presume that 3g KWG is the optimal dose where greatest improvements on Al were seen, while the 1g and 6g KWG are containing either low or high levels of ginsenosides in order to elicit favorable responses, being outside the therapeutic dose range. This in turn underscores the importance of identifying an efficacious range of ginsenoside levels so as to attain sustainable and reproducible ameliorative effects. From this, and as previously observed in similar study designs\(^{109,213}\), we learn that higher amounts of ginsenoside concentrations are not necessarily associated with greater reductions on either postprandial glycemic or vascular parameters, all of which supports the concept of a therapeutic dose range for ginsenosides.
As well, the significant differences observed with both the 3g and 1g KWG doses compared to the 6g KWG dose at the 4hr time point is in line with the suggestion that low or moderate doses rather than high doses of ginsenosides may improve vascular function. Such an observation is not atypical for the ginsenosides as we previously showed that low levels of ginsenosides from KRG body excreted significant BP reductions, while lack of a BP-reducing effect was associated with higher ginsenoside levels from KRG rootlets \(^{213}\). Support for this also comes from animal data where Kaku et al. \(^{270}\) found depressor effects following low-dose ginsenoside injections to rats, as well as pressor effects with the high-dose injections. Additionally, Chen et al. \(^{271}\) showed that ginsenosides were associated with both dilation and constriction effects in isolated arteries, where a dilation effect prevailed at low doses, and a balance between the two processes occurred at high doses. Collectively, data from the literature is in support of the possibility that low or moderate doses of ginsenosides may improve vascular function, as observed with the 3g KWG, while high doses may not, as seen with the 6g KWG.

While previous findings have suggested that Rg3, a ginsenoside generated during steam processing that is unique to KRG, exhibits vasodilatory potential \(^{143,201,207,210}\), and is regarded as one of the most potent ginsenosides \(^{210}\), our favorable vascular findings herein, with the non-steamed ginseng variety, may provide additional insight into the potential constituents that are responsible for the vascular improvements seen with 3g KWG intervention. Further expanding on this, accumulating data from animal and in vitro work have demonstrated that certain ginsenosides, such as Rb1, Re, and Rg1, display cardioprotective effects, chiefly on vascular endothelial function via endothelium dependent release of NO \(^{173,226,272}\). In view of the clinical implications linking arterial stiffness and endothelial dysfunction to decreased NO generation and increased NO inactivation \(^{273,274}\), together with evidence from preclinical data, the beneficial AI findings seen with 3g KWG may be possibly explained by NO driven mechanism stimulated by these ginsenosides, including Rb1, Re, or Rg1, two of which (Re and Rg1) were found to be
present in fairly similar concentrations as in our efficacious KRG roots. While it is tempting to attribute our favorable vascular observations to these ginsenosides, the possibility that other potential active fractions of ginseng, including unmeasured ginsenosides such as the malonyl ginsenosides, polysaccharides, peptides, fatty acids, and polyacetylenic alcohols might have played a role, cannot be eliminated.

The lack of a significant amelioration in peripheral systolic BP is not unexpected and is partly in line with our previous observations, where KRG showed either neutral or moderate effects. Given the notion that certain vasodilatory agents may exert a significant lowering effect on ventricular workload by decreasing muscular artery stiffness and reflected wave amplitude, without a direct effect on elastic arteries and reduction in systolic blood pressure, it can be implied that improvements in arterial wave reflection, or AI, may occur independent of any possible effects on BP measures. Although KWG did not lower BP relative to control, the neutral BP observations are significant in light of the concern in the literature that ginseng may increase blood pressure, which resulted in advice given to individuals with hypertension to avoid ginseng products. Thus, this study adds further information on the safety of ginseng in hypertension.

5.6 Limitations

Two limitations to this work must be addressed. First, we assessed the vascular and glycemic health effects of KWG in an acute design. It is thus unclear whether continuous long term administrations would yield similar results. Secondly, the subjects were taking diabetes and antihypertensive medications. Though these medications were not taken on morning visits, they were taken 12-24 hrs prior to the visits. Hence, they or their metabolites might have been present in the blood on test mornings, and could have potentially confounded the participants’ vascular and glycemic measures.
5.7 Conclusions

In conclusion, the current study is the first to explore the glycemic and vascular effects of KWG in subjects with T2DM. It showed that acute administration of 3g KWG elicited modest but significant reductions in arterial stiffness, as measured by AI, while glycemia was not affected. Interestingly, the optimal dose where greatest improvements were seen across glycemia and vascular parameters appears to be the 3g dose which were not observed with neither the 1g nor the 6g dose. In light of such findings, future studies should aim at exploring the long term vascular and glycemic effects of the optimal dose identified herein, 3g, with a main focus in determining its contributing constituents. Should the beneficial results on AI observed in this study be validated in longer term trials, the 4-yr old KWG root may be a promising candidate that is comparable in efficacy to the typically endorsed and valued 6-yr old KRG, creating significant economic possibilities in the area of evidence-based remedies for CVD risk reduction.
CHAPTER 6

OVERALL DISCUSSION, STRENGTHS, AND LIMITATIONS
6. OVERALL DISCUSSION, STRENGTHS, AND LIMITATIONS

6.1 Overall discussion

This thesis explored the glycemic and vascular health benefits of ginseng through the conduction of two projects; a systematic review and meta-analysis, and an acute, randomized, double blind, placebo-controlled, clinical trial. The systematic review and meta-analysis showed that ginseng intervention had modest, yet significant, lowering effects on FBG relative to control in subjects with and without diabetes (MD=-0.31 mmol/L; P=0.03), with subgroup analysis further demonstrating greater FPG reductions in those with diabetes than those without. This ameliorative effect, however, was not observed on FPI, HbA1c, or HOMA-IR. Additionally, though ginseng intervention did not demonstrate significant reductions on the long term marker of glycemic control, HbA1c, subgroup analyses showed significant reductions in HbA1c in parallel compared to crossover trials (P=0.01).

Findings from the dose-response acute RCT in subjects with T2DM revealed that KWG does not improve any glycemic or BP parameters when compared to control, irrespective of the dose administered. However, significant improvements in the secondary outcome measure, arterial stiffness, a validated measure of overall vascular health, as measured by AI were seen with 3g KWG dose (MD=-3.30%; P=0.04). It is noteworthy that the benefits seen on AI occurred despite the participants’ well controlled glucose and BP levels, as a result of early stage of disease or/and extensive medication use. Thus it becomes a very significant finding that KWG can cause further improvement in arterial compliance, which is often difficult to achieve with added therapy. Moreover, given that a dose-response exploration was conducted for determination of a potential optimal dose, which can be ultimately used in long term investigation, our results should be interpreted within this context.
Establishing an effective therapeutic dose range in pharmacological applications is of pivotal importance, as doses beyond the established efficacious range can be considered noxious or have contradictory effects. We show that while no significant reductions on PPBG were observed with any of the KWG doses relative to control, a U-shaped dose-response relationship was seen after single bolus administrations of escalating KWG quantities. The data indicates that 1g KWG is presumably not delivering sufficient amount of bioactives, or ginsenosides, in order to elicit a glycemic response, while a load of 6g KWG has possibly reached a point of substrate saturation, where increases in PPBG are observed, though not significantly different from control, as similarly observed in our previous work\textsuperscript{109,186}. Although 3g KWG showed the greatest reduction across both PPBG iAUC and PPBG measures, these reductions were not significant when compared to control. Yet, despite these insignificant findings, the potential for KWG to benefit glycemia cannot be exclusively disregarded due in large part to the presence of possible confounders that might have precluded accurate assessment of KWG’s effect on blood glucose. Similarly, the acute dose response observations on AI at 240 minutes add to our glycemic findings further supporting the concept of an efficacious therapeutic dose range for ginsenosides. The efficacy of the KWG doses seen with the AI findings are in accordance with our glycemic observations, where 3g KWG appears to exhibit an optimal level of ginsenosides rendering a pronounced lowering effect, while the levels of ginsenosides in the 1g and 6g KWG doses appear to fall outside the efficacy range suggesting that either insufficient levels of ginsenosides are present, as with the 1g KWG, or the possibility of a saturation-limiting activity of ginsenosides, as with the 6g KWG. This in turn adds support for the suggestion that low or moderate levels rather than high levels of ginsenosides may be more efficacious in eliciting favorable vascular effects, as seen in previous findings\textsuperscript{213}. 
In light of the well established phenomenon that higher levels of baseline glycemia would result in greater reductions with anti-hyperglycemic agents\textsuperscript{61}, and given that HbA1c is a component of both the FBG and PPBG, where correlations between PPBG and HbA1c values are seen best when levels are approaching the target value of ≤7.0%\textsuperscript{266-268}, it can be postulated that the insignificant glycemic findings seen with 3g KWG might possibly be interpreted by the strictly controlled glycemic status of our participants (HbA1c=6.9%), resulting from their use of anti-diabetic agents. Further support to this can be provided by our meta-analysis results were participants with diabetes exhibited greater reductions in FBG than those without, as well as by our continuous meta-regression analyses, which demonstrated that increases in baseline FBG were linearly associated with FBG reductions on the ginseng interventions. Collectively, our results indicate that ginseng can depict greater glycemic benefits in individuals with compromised glycemic health. Accordingly, the clinical implications of our findings may still be relevant, and may potentially be affirmed with future investigations. Based upon the clinical findings, we can hypothesize that 3g KWG is the optimal dose with potential clinical implications, as it generated the greatest improvements and did so consistently across both PPBG and AI parameters.

Taken as a whole, findings from our meta-analysis suggest that ginseng may improve glycemic control when used as an adjunct to conventional diabetes therapy, where greater glycemic improvements were observed in individuals with increased glycemic distortions. Our clinical trial results further indicate that the non-steamed Korean ginseng variety, that is KWG, may exhibit favorable vascular health benefits as demonstrated by improvements in AI. These preliminary findings indicate that KWG might be a promising therapeutic agent for improving certain CVD risks, where its efficacy could parallel that of the highly regarded KRG root.
6.2 Strengths

6.2.1 Systematic review and meta-analysis:

Strengths of the systematic review and meta-analysis projects include:

(1) Only RCTs were included in the analysis, which are considered the gold standard for measuring an intervention's impact on a certain outcome/parameter. Persuasive evidence suggests that they are superior to other trial designs in estimating an intervention’s true effect due but not limited to the following: a) Randomization of study interventions reduces the possibility of selection bias; b) Blinding of participants and/or observers is maintained which protects against allocation bias by the study investigator, and increases the credibility of the results; c) The study intervention is compared to a control group, which provides a base upon which the effects of the study intervention can be assessed.

(2) The literature search was not restricted to a specific language, allowing for a more comprehensive inclusion of trials.

(3) Publication bias was examined by both visual inspection of funnel plots, as well as Egger’s and Begg’s tests, providing a more accurate assessment of the presence of any potential bias in the publication of studies showing improvements with the ginseng intervention with respect to glycemic control.

(4) Trials that assessed ginseng as part of a multi-herbal treatment were excluded so as to isolate the effect of ginseng from other components that might be present in the herb.

6.2.2 Randomized clinical trial

Strengths of the RCT include:

(1) The randomized, double-blind, placebo-controlled design of the trial offers the gold standard for assessing the effectiveness and safety of a certain intervention, as described in section 6.2.1.
(2) The crossover design of the trial reduces the between-patient variation by allowing each subject to act as his or her own control. It also requires fewer patients than a parallel study design for an equal number of treatment comparisons, because each experimental unit (patient) can be used several times.

(3) The wheat bran control was administered on two occasions, and averaged, allowing for a truer estimation of the control.

(4) Based on power analysis, the sample size of the trial was sufficient to detect meaningful postprandial blood glucose effects of the KWG interventions relative to control.

6.3 Limitations

6.3.1 Systematic review and meta-analysis

Limitations of the systematic review and meta-analysis project include:

(1) The effect of ginseng dose was not evaluated though it was specified a priori. This was largely owed to the variation in ginseng preparations administered (whole root/rootlets of Korean red/Panax/American ginseng vs. often uncharacterized extracts of these varieties), precluding calculation of ginseng dose equivalents. As a result, this made the pooling of doses and comparing their effects between trials rather difficult.

(2) Most of the trials were fairly of short duration, with only 3 out of the 9 trials that assessed HbA1c having a follow up duration of ≥12 weeks. Consequently, this might have possibly led to misleading conclusions regarding ginseng’s potential to ameliorate long term glycemic control.

(3) Several trials included subjects receiving either insulin or oral hypoglycemic medications which might have affected their glycemic measures, and thus, the true hypoglycemic potential of ginseng might have been underestimated.
(4) Variability in ginsenoside composition coupled with poor standardization of the ginsenoside profile complicates the assessment of ginseng’s glycemic benefits, particularly in light of evidence showing that the anti hyperglycemic efficacy of ginseng might be as highly variable as its ginsenoside composition252.

(5) Most trials did not provide information on ginsenoside profile, making it hard to verify whether the ginsenoside profile was similar across studies that showed effective results.

(6) Given that the anti-diabetic effects of ginseng was demonstrated throughout different ginseng species, doses, and study populations, data cannot be regarded as highly robust considering the evident pharmacological differences seen among different ginseng species, batches, and preparations277.

(7) Unexplained heterogeneity was observed in the overall analysis, albeit accounting for this heterogeneity. Collectively, these limitations reinforce the need for longer, better designed clinical trials that report data more systematically and transparently to better understand the effect of ginseng on glycemia.

6.3.2 Randomized clinical trial

Limitations of the RCT include:

(1) The acute design of the study creates uncertainty as to whether improvements in arterial stiffness will persist if KWG is administered on a long-term basis.

(2) Our study population was comprised of individuals on anti-diabetic and anti-hypertensive agents that were consumed 12-24 hrs prior to the start of study. These agents might have presented themselves as confounding factors as they could have residually affected their glycemic and BP measures, and thus, an accurate assessment of KWG’s effect on glycemic and vascular parameters might have been complicated.
6.4 Clinical implications

It has become well known that optimal glycemic control is fundamental in diabetes management\(^4\). Despite the clinical trial not showing significant reductions in PPBG iAUC or PPBG with escalating doses of KWG compared to control, the systematic review and meta-analysis did find significant modest reductions in FBG with ginseng intervention relative to control \((P=0.03)\), along with greater FBG reductions in those with than without diabetes. To extend from this, our \textit{a priori} subgroup analysis further demonstrated significant benefit \((\sim 0.2\% \text{ reduction})\) in the long term marker of glycemic control, HbA1c, in parallel versus crossover design trials \((P=0.01)\). The benefit in people with diabetes was seen beyond that of the oral anti-hyperglycemic medications that they were using, implying that ginseng may have advantages as an adjunct to conventional therapy. Yet, the lack of an overall amelioration in HbA1c raises some doubts as to the long-term being drawn at this time.

Augmentation index, the method by which arterial stiffness was assessed in the current trial, is a measure of arterial wave reflection generated via pulse wave analysis. It provides significant information on systemic arterial stiffness and workload of the heart\(^93\). Increases in AI imply augmented wave reflections from the periphery or a faster return of the reflected wave as a result of increased PWV. Enhanced wave reflections, in turn, increase left ventricular load and impair ventricular function\(^278\). In light of the additional prognostic information provided by Al concerning CVD risk, it has become increasingly recognized as a valuable marker and independent predictor of CVD morbidity and mortality\(^84\). Given that increased arterial stiffness is demonstrated in individuals with diabetes\(^279\), treatments should aim at reducing arterial stiffness in order to lower afterload and pulse pressure, and promote regression of left ventricular and arterial wall hypertrophy. Accordingly, the acute improvements seen with the 3g KWG on AI may have significant clinical implications, where it may offer a NHP alternative to numerous
pharmacological agents that have shown to reduce arterial stiffness, and consequently, present itself as a promising therapeutic agent for improving certain CVD risks. However, despite these acute promising results, and before any assertions can be made, long term assessment of the effect of 3g KWG on AI is necessary.

6.5 Future Directions

Future investigations should aim at addressing the limitations of this thesis. In light of the continuous heterogeneity observed in the meta-analysis project, and in order to provide more precise estimates of ginsengs long-term effectiveness, longer-term, well-powered, RCTs of the effect of established ginseng preparations on glycemic parameters using intention-to-treat analyses are warranted. In the absence of standardized products on which to perform trials, there is also an imperative need to develop a basis for ginseng standardization in diabetes by investigating the compositional features which explain the glycemic-lowering effects of ginseng, taking into account unexamined non-saponin components such as the polysaccharides, panaxosides, and quinquefolans.

As for the clinical trial findings, the results obtained must be ultimately validated in a longer-term trial in order to: (1) Ensure that the effects found from single administration are confirmed; and (2) Evaluate long term effects which cannot be observed in a single dose administration. Active components, ginsenosides are steroidal saponins that, based on their structure, are postulated to have some effect on gene transcription, in which case longer term administration in humans is the only way to provide answers on these potential benefits. A recommendation following the results observed here is to undertake a clinical trial of 3-6 months duration at minimum using the most optimal dose identified in this trial of 3g. Moreover, the study population should include those with T2DM, who exhibit compromised metabolic functions, but are not treated with anti-diabetic and anti-hypertensive medications so as to prevent their potential
confounding effects on both glycemia and BP measures. Accordingly, should the reductions observed herein be sustained in longer term investigations, it would generate findings that are of significant clinical benefit, and would further help to ultimately develop a clinically-validated, economical alternative therapy that can be used in the treatment of diabetes and related CVD risks worldwide.

6.6 Conclusions

There is a promising potential role for ginseng in modestly improving glycemic and vascular health parameters when taken as an adjunct to conventional medicine.

This thesis work illustrated the following:

1. In a systematic review and meta-analysis of 16 RCTs (n= 770), ginseng intervention modestly, yet, significantly lowered FBG compared to control (MD= -0.31 mmol/L [95% CI: -0.59, -0.03]; P=0.03). No significant improvements with the ginseng intervention were observed on HbA1c, FPI, and HOMA-IR relative to control.

2. In a double-blind, randomized, placebo-controlled, crossover trial in 25 participants with well-controlled T2DM, acute administration of escalating doses of KWG was safe, but did not significantly improve postprandial glycemic and BP measures including PPBG iAUC, PPBG, CBP, and BBP relative to control. However, 3g KWG did show significant reductions (MD= -3.30%; P=0.04) on AI, a marker of arterial stiffness, when compared to control.

To conclude, this thesis work substantiates the ameliorating effects of ginseng on both glycemic and vascular parameters, with a focus on the rarely studied, yet widely consumed, non-steamed Korean ginseng variety, KWG. It also highlights the imperative need for better standardization of ginseng’s active components in order to ensure efficacy, reproducibility, and product quality.
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CONSENT TO PARTICIPATE IN A RESEARCH STUDY

TITLE OF RESEARCH STUDY

Acute Dose Response effects of Korean White ginseng (Panax Ginseng C.A. Meyer) on Cardiovascular Disease Risk Factors in Individuals with Metabolic Syndrome or type 2 diabetes: Pilot Double Blind, Randomized, Placebo-controlled trial

PRINCIPAL INVESTIGATOR/SPONSOR
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Ms Esra Shishtar, MSc Candidate, is involved in this study as part of her education to complete a Masters degree at the Department of Nutritional Sciences, University of Toronto, thus she also has an academic interest in completing this study. Ms Shishtar’s faculty supervisor is Dr. Vladimir Vulcan.

FUNDING SUPPORT
Department of Herbal Crop Research;
National Institute of Horticultural & Herbal Science, Rural Development Administration (RDA), Korea

STUDY PRODUCTS
Korean White Ginseng: provided by Department of Herbal Crop Research; National Institute of Horticultural & Herbal Science, Rural Development Administration (RDA), Korea
Korean Red Ginseng: provided by BTGin Ltd. (Daejeon, Korea)
Wheat Bran: provided by Department of Herbal Crop Research; National Institute of Horticultural & Herbal Science, Rural Development Administration (RDA), Korea
Study Capsules: provided by Department of Herbal Crop Research; National Institute of Horticultural & Herbal Science, Rural Development Administration (RDA), Korea; provided by BTGin Ltd. (Daejeon, Korea)

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KWG Study, Consent Form, Version Date: 08 April 2013

Participant's Initials: ______
STATEMENT OF CONSENT

TITLE OF RESEARCH STUDY

Acute Dose Response effects of Korean White ginseng (Panax Ginseng C.A. Meyer) on Cardiovascular Disease Risk Factors in Individuals with Metabolic Syndrome or type 2 diabetes: Pilot Double Blind, Randomized, Placebo-controlled trial

The research study has been explained to me, and any questions that I had have been answered to my satisfaction. I have been informed of the alternatives to participation in this study. I have the right not to participate and the right to withdraw without affecting the quality of medical care at St. Michael’s Hospital for me and for other members of my family. As well, the potential harms and benefits (if any) of participation in this research study have been explained to me.

I have been told that I have not waived my legal rights nor released the investigators, sponsor, or involved institutions from their legal and professional responsibilities. I know that I may ask now, or in the future, any questions I have about the study. I have been told that records relating to me will be kept confidential and that no information will be disclosed without my permission unless required by law. I have been given sufficient time to read the above information.

Please check the appropriate box, and initial, to indicate your selection:

☐ (Initials): YES, I agree that you may inform my primary care/treating physician in writing of my results in this study.

☐ (Initials): NO, I do not agree that you may inform my primary care/treating physician in writing of my results in this study; or NO, I do not have a primary care/treating physician.

I hereby consent to participate in this study. I will be given a signed copy of this consent form.

Name of Participant (print): ________________________________

Signature of Participant: ________________________________ Date: ________________________________

Name and Position of Person Obtaining Consent (print): ________________________________

Signature of Person Obtaining Consent: ________________________________ Date: ________________________________

Page 8 of 8
KWG Study, Consent Form, Version Date: 08 April 2013

Participant’s Initials: ___
Appendix 2. St. Michael's Hospital Research Ethics Board Original Approval

Research Ethics Office
Telephone: (416) 954-4000 Ext. 2557
Fax: (416) 954-3343
E-mail: rebo@smah.toronto.on.ca

April 12, 2013

Dr. Vladimir Vuksan,
Division of Endocrinology and Metabolism,
Diabetes Comprehensive Care Program,
St Michael's Hospital

Dear Dr. Vuksan,

Re: REB# 12-300 - Acute Dose Response effects of Korean White ginseng (Panax Ginseng C.A. Meyer) on Cardiovascular Disease Risk Factors in Individuals with Metabolic Syndrome or type 2 diabetes: Pilot Double Blind, Randomized, Placebo-controlled trial

REB APPROVAL: Original Approval Date April 12, 2013
Annual/Interval Review Date April 12, 2014

Thank you for your application submitted on 09 October, 2012. At the St Michael's Hospital (SMH) Research Ethics Board (REB) meeting held on November 07, 2012, the above referenced study was discussed and subsequently the views derived from this discussion have been documented and resolved.

The REB approves the study as it is found to comply with relevant research ethics guidelines, as well as the Ontario Personal Health Information Protection Act (PHIPA), 2004. The REB hereby issues approval for the above named study for a period of 12 months from the date of this letter. Continuation beyond that date will require further review of REB approval. In addition, the following are appropriate and hereby approved:

5. Newspaper Ad (text submitted 11 April 2013)
6. Telephone Script, Version Date: 29 November 2012 (submitted 29 November 2012)
7. Consent Form, Version Date: 08 April 2013
8. Letter of Appreciation, Ver.2 04/09/13 (clean copy, submitted 09 April 2013)

Furthermore, the following documents have been received and are acknowledged:


Dr. Vladimir Vuksan (REB# 12-300) REB Letter April 12, 2013
5. Information/Medical History Form, Ver.2 11/29/12  
   (clean copy, submitted 29 November 2012)  
6. Preclinical Information [for Screening visit] Ver.1 04/09/13  
   (clean copy, submitted 09 April 2013)  
7. Preclinical Information [for Study Dosing visits] Ver.4 04/11/13  
   (clean copy, submitted 11 April 2013)  
8. Clinical Assessment, Ver.4 04/09/13  
   (clean copy, submitted 09 April 2013)  
9. Dosing Assignment Form  
   (clean copy, submitted 11 April 2013)  
10. Safety Questionnaire, Ver.4 04/09/13  
    (clean copy, submitted 09 April 2013)  
11. Symptoms Questionnaire, Ver.4 04/09/13  
    (clean copy, submitted 09 April 2013)  
12. 24 Hour Symptoms Assessment Form, Ver.3 04/09/13  
    (clean copy, submitted 09 April 2013)  
13. Telephone Follow-up Symptoms Form, Ver.3 04/09/13  
    (clean copy, submitted 09 April 2013)  
14. Serious Adverse Events Reporting Form, Ver.2 04/09/13  
    (clean copy, submitted 09 April 2013)  

Please also note that Dr. Amir Hanna, although a member of the St. Michael's Hospital Research Ethics Board, did not have a role in the deliberation, review or approval of this study.

During the course of this investigation, any significant deviations from the approved protocol and/or unanticipated developments or significant adverse events should immediately be brought to the attention of the REB.

Please note that if a Clinical Trial Agreement is required, it must be submitted to the Office of Research Administration for review and approval. Any additional institutional approvals must be coordinated and approved through the Office of Research Administration prior to initiation of this research. All drug dispensing must be coordinated through the Research Pharmacy at 416-864-5413.

The St. Michael's Hospital (SMH) Research Ethics Board (REB) operates in compliance with the Tri-Council Policy Statement Ethical Conduct for Research Involving Humans, the Ontario Personal Health Information Protection Act, 2004, and ICH Good Clinical Practice Consolidated Guideline E6, Health Canada Part C Division 5 of the Food and Drug Regulations, Part 4 of the Natural Health Product Regulations, and the Medical Devices regulations. Furthermore, all investigational drug trials at SMH are conducted by Qualified Investigators (as defined in the latter document).

With best wishes

☐ Dr. Bob Hyland  
Chair, Research Ethics Board  

☑ Dr. Brenda McDowell  
Vice Chair, Research Ethics Board
Appendix 3. Health Canada Notice of Authorization

September 5, 2012

Dr. Vladimir Vukman (c/o Elena Jovanovski)
Risk Factor Modification Centre, St. Michael’s Hospital
PO Box 30 Bond Street, 19th Floor, Queen Wing
Toronto, Ontario
M5B 1W8

Dear Dr. Vukman:

Re: CLINICAL TRIAL APPLICATION for [Korean White Ginseng (Panax ginseng C.A. Meyer)]
Natural Health Products Regulations; Section 61

The Natural Health Products Directorate, Bureau of Clinical Trials and Health Sciences, is pleased to inform you that the information and material provided to support the above Clinical Trial Application, have been assessed and we have no objection to your proposed study. Please consider this as your notice of authorization to sell or import this natural health product for the purposes of this clinical trial in Canada.

I would remind you of the necessity of complying with the Natural Health Products Regulations, Part 4, in the sale of this product for clinical setting. In addition, the Regulations (Part 4) impose responsibilities, including commencement notice, record keeping and reaction reporting, on those conducting clinical trials. Please ensure that all systems are compliant in order to meet these responsibilities.

To notify NHPD in an expedited manner in the case of serious adverse reactions and/or serious unexpected adverse reactions, please fax your report(s) to the following number: 613-946-0174.

You are also reminded that all clinical trials should be conducted in compliance with the Health Canada Guidance for Industry: Good Clinical Practice: Consolidated Guidelines ICH Topic E6.

Should you have any questions concerning this letter, please contact the submission coordinator at nhpdcda.dec-dpca@hc-sc.gc.ca.

Yours sincerely,

[Signature]

Adam Gibson
Senior Executive Director, Bureau of Product Review and Assessment
Natural Health Products Directorate
2936 Baseline Rd. (A.L. 3302C), Ottawa, ON K1A 0E9

Canada
### Preclinical Information

*Date: ___________________________
TO BE COMPLETED BY THE SUBJECT*

| Did you consume at least 150g (6 oz.) of carbohydrate on each of the two days previous to this test? This amount is equivalent to 3 servings of any of the following alone or in combination: 2 slices of bread, 1 cup of cooked rice/pasta, 1 medium potato, 1 bowl of cereal with milk, 1 glass of juice/soft-drink, 3 oranges/apples, or 1 bowl of ice cream? □ Yes  □ No
| If you consumed any of the foods listed below yesterday at any time please circle the appropriate food:
| Potato-chips  Pretzels  Crackers  Fast Foods  Olives  Pickles  Sauerkraut  Soy Sauce  Ketchup  Cheese  Canned Soup  Cake/Pastries  Bouillon  Ham  Sausages  Hot-Dog  Smoked Meat  Sardines  Anchovies  Tomato Juice (can)  Frozen Vegetables  Canned Vegetables
| Did you take any medications (prescription, OTC, etc.), or supplements last night or this morning? If yes, please describe. □ Yes  □ No
| Describe:
| Type:
| Did you do anything since your last visit that is not part of your regular routine? This may include exercise or use of alcohol, medications, or supplements. If yes, then please describe. □ Yes  □ No
| Describe:
| How long ago did you last (1) empty your bladder and/or (2) have a bowel movement? (1) Last urination _____ hour(s) ago (2) Last bowel movement _____ hour(s) ago
| How many hours of sleep did you have last night? Does this represent a typical amount? □ Yes  □ No  ________ hours
| Did you do anything before the test that is not in compliance with the study or part of your regular routine? If yes, then please describe. □ Yes  □ No
| Describe:
| What was your mode of transportation to the clinic this morning? Is this different from other clinic mornings? □ Yes  □ No
| Comment:
| How would you rate your current level of health/well-being? Please comment on anything unusual. □ Excellent  □ Good  □ Fair  □ Poor

*P.I Dr. Vukan, V. KWG in Type 2 diabetes or Metabolic syndrome  Ver.1 04/09/13*
Appendix 5. Clinical Assessment Form

CLINICAL ASSESSMENT

Date: ___________ Visit Number: 1 2 3 4 5 6 (Circle)

TO BE COMPLETED BY THE INVESTIGATOR

BASELINE MEASUREMENTS

Anthropometry & blood pressure measures

Ht (cm):__________

Wt (kg):__________ BMI (wt/h²):__________

BF (%):__________ (Attach TANITA printout)

Brachial blood pressure

SBP/DBP/HR (mmHg):

Avg: ___/___/___

1. ___/___/___

2. ___/___/___

3. ___/___/___

Central blood pressure (mmHg):

1. ___/___/___

2. ___/___/___

3. ___/___/___

Capsule consumption time (12 Capsules with 50g glucose load): __________ min

Capillary blood sample & vascular measures

Capillary baseline blood sample (mmol/L):

Arterial Stiffness results:

Aix (%): 1: ____ 2: ____ 3: ____

SEVR: _______ _______ _______

ED (sec): _______ _______ _______

24-hr ABP (SBP/DBP) (mmHg): attached

Yes____ No____

POST-TREATMENT MEASUREMENTS

Capillary blood samples (mmol/L):

<table>
<thead>
<tr>
<th>Time (Minutes)</th>
<th>15’</th>
<th>30’</th>
<th>45’</th>
<th>60’</th>
<th>90’</th>
<th>120’</th>
<th>180’ Standardized Breakfast Meal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Capillary blood sample (mmol/L)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

P.I. Dr. Vukasin, Y. KWG in Type 2 diabetes or Metabolic syndrome Ver.4 04/09/13
### Vascular measures:

<table>
<thead>
<tr>
<th></th>
<th>60°</th>
<th>120°</th>
<th>180° Standardized Breakfast Meal</th>
<th>240°</th>
</tr>
</thead>
<tbody>
<tr>
<td>Augmentation Index (Alx) (%)</td>
<td>1:</td>
<td>2:</td>
<td></td>
<td>3:</td>
</tr>
<tr>
<td>Central BP (mmHg)</td>
<td>1:</td>
<td>2:</td>
<td></td>
<td>3:</td>
</tr>
<tr>
<td>SEVR</td>
<td>1:</td>
<td>2:</td>
<td></td>
<td>3:</td>
</tr>
<tr>
<td>ED (sec)</td>
<td>1:</td>
<td>2:</td>
<td></td>
<td>3:</td>
</tr>
<tr>
<td>Brachial blood pressure (done at 60°, 120°, and 180°)</td>
<td>1:</td>
<td>2:</td>
<td></td>
<td>3:</td>
</tr>
<tr>
<td>SBP/DBP/HR (mmHg)</td>
<td>1:</td>
<td>2:</td>
<td></td>
<td>3:</td>
</tr>
</tbody>
</table>
# Symptoms Questionnaire

To be completed by the participant

Visit Number: 1 2 3 4 5 6 (Circle)

Please indicate if you have any of the following symptoms during the study visit:

<table>
<thead>
<tr>
<th>Symptoms</th>
<th>Presence</th>
<th>Severity</th>
<th>Time</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bloating</td>
<td>□ Yes</td>
<td>Low</td>
<td>1-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>□ No</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Belching</td>
<td>□ Yes</td>
<td>Low</td>
<td>1-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>□ No</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Diarrhoea</td>
<td>□ Yes</td>
<td>Low</td>
<td>1-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>□ No</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Flatulence (gas)</td>
<td>□ Yes</td>
<td>Low</td>
<td>1-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>□ No</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Excessive urination</td>
<td>□ Yes</td>
<td>Low</td>
<td>1-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>□ No</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nausea</td>
<td>□ Yes</td>
<td>Low</td>
<td>1-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>□ No</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Headache</td>
<td>□ Yes</td>
<td>Low</td>
<td>1-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>□ No</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dizziness</td>
<td>□ Yes</td>
<td>Low</td>
<td>1-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>□ No</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Disorientation</td>
<td>□ Yes</td>
<td>Low</td>
<td>1-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>□ No</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nervousness (Anxiety)</td>
<td>□ Yes</td>
<td>Low</td>
<td>1-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>□ No</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>□ Yes</td>
<td>□ No</td>
<td>Low 1—2—3—4—5—6—7—8—9—10 High</td>
<td></td>
</tr>
<tr>
<td>----------------</td>
<td>------</td>
<td>------</td>
<td>--------------------------------</td>
<td></td>
</tr>
<tr>
<td><strong>Insomnia</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Numbness</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Impaired vision</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Other</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(specify):</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
## 24 Hour Symptoms Assessment Form

**To be completed by the participant**

Date: ________________  Visit: 1  2  3  4  5  6 (circle)

Please indicate if you have experienced any of the following symptoms in the 24-hours after your visit. Please bring back this form completed on your next scheduled visit. Thank you.

<table>
<thead>
<tr>
<th>SYMPTOMS</th>
<th>PRESENCE</th>
<th>ONSET / DURATION</th>
<th>Comment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bloating</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Belching</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Diarrhea</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Flatulence (gas)</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Nervousness (anxiety)</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Nausea</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Headache</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Dizziness</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Disorientation</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Insomnia</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Excessive urination</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Other: (specify)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Appendix 8. Telephone Follow-up Symptoms Form

**Telephone Follow-up Symptoms Form**

To be completed by the investigator

Date: ____________, Visit # 6

Begin by saying: I’m calling to follow up with you and inquire if you have experienced any symptoms in the 24-hours after your last visit to the Risk Factor Modification Center. After that, go through each symptom and ask if they had experienced it: (e.g., did you feel any bloating or belching? How about diarrhea? etc…)

<table>
<thead>
<tr>
<th>SYMPTOMS</th>
<th>PRESENCE</th>
<th>ONSET / DURATION</th>
<th>Comment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bloating</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Belching</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Diarrhea</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Flatulence (gas)</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Nervousness (anxiety)</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Nausea</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Headache</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Dizziness</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Disorientation</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Insomnia</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Excessive urination</td>
<td>□ Yes</td>
<td>□ No</td>
<td></td>
</tr>
<tr>
<td>Other symptoms? Specify?</td>
<td>________</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
## Appendix 9. Medical Information Form

### INFORMATION/MEDICAL HISTORY FORM

All information provided in this questionnaire will be kept confidential and released only for the purpose of the present study.

<table>
<thead>
<tr>
<th>Gender:</th>
<th>Waist circumference (cm):</th>
<th>Brachial blood pressure (mmHg):</th>
</tr>
</thead>
<tbody>
<tr>
<td>□ Male</td>
<td>BF (%): _________________</td>
<td>_______________________________</td>
</tr>
<tr>
<td>□ Female</td>
<td>_________________________</td>
<td>_______________________________</td>
</tr>
</tbody>
</table>

If needed:

| HDL-C (mg/dL): | _________________________ |
| TG (mg/dL): | _________________________ |
| HbA1c (%): | _________________________ |

Capillary blood glucose (mmol/L):

Urine pregnancy test: (P) / (N)

### High blood sugar

Has your doctor ever told you that you have high blood sugar, high blood pressure? If yes, then please give details: when, how high, medications (Rx), complications, etc. □ Yes  □ No

<table>
<thead>
<tr>
<th>When:</th>
<th>How high:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Fasting glucose: __________ mmol/L</td>
</tr>
<tr>
<td></td>
<td>Post-meal glucose: __________ mmol/L</td>
</tr>
<tr>
<td></td>
<td>HbA1c (glycosylated haemoglobin): __________ %</td>
</tr>
<tr>
<td>Rx:</td>
<td>_________________________</td>
</tr>
</tbody>
</table>

Complications: _________________________

### High blood pressure

<table>
<thead>
<tr>
<th>When:</th>
<th>How high:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>sBP/dBP: __________ / __________ mmHg</td>
</tr>
<tr>
<td>Rx:</td>
<td>_________________________</td>
</tr>
</tbody>
</table>

Complications: _________________________

### Does anyone in your family have diabetes, high blood pressure, or heart disease? If yes, then please describe, indicating how long they have had it and their relationship to you. □ Yes □ No

<table>
<thead>
<tr>
<th>□ Mother</th>
<th>□ Father</th>
</tr>
</thead>
<tbody>
<tr>
<td>□ Siblings</td>
<td>□ Aunt/Uncle</td>
</tr>
<tr>
<td>□ Grandmother/grandfather</td>
<td>□ Grandmother/grandfather</td>
</tr>
</tbody>
</table>

### Do you take medications, herbs or supplements? If yes, then please describe, indicating types, brand names, doses, and times. □ Yes □ No

Describe: _________________________

---

*P.J Dr. Yuksan, V. KWG in Type 2 diabetes or Metabolic syndrome  Ver.2 11/29/12*
<table>
<thead>
<tr>
<th>CONDITION</th>
<th>NO</th>
<th>YES</th>
<th>Onset date</th>
<th>Present status</th>
<th>Active (please indicate treatment)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Malabsorption syndrome</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Crohn’s disease</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Celiac disease</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ulcerative colitis</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stomach (gastric) ulcer</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Duodenal ulcer</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Intestinal parasites</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Diarrhea (&gt; 2 liquid stools/day)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Constipation (≥ 3 days duration)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heart disease</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stroke</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heart attack</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Arrhythmia</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uncontrolled hypertension</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Systolic BP ≥ 140</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Diastolic BP ≥ 90</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Blood clotting disorders</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anaemia</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kidney disease</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*P.I Dr. Vuksan, V. KWG in Type 2 diabetes or Metabolic syndrome*  
*Ver. 2 11/29/12*
<table>
<thead>
<tr>
<th>Liver disease</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Infectious hepatitis (B, C, D)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Recently diagnosed infectious hepatitis A, E</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Retinopathy (persistent or acute damage to the retina of the eye)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HIV/ AIDS</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tuberculosis</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cancer</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thyroid disease</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Do you experience any of the following:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fatigue</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unexplained weight gain</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dry skin and hair</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Depressed mood</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cold intolerance</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Constipation</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Increased cholesterol?</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nervousness/irritability</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Palpitations</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heat intolerance</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Increased sweating</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unexplained weight loss</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Insomnia</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pancreatic disease</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Diabetes</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Asthma</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Any food allergies</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Allergies to ginseng, wheat bran, or gelatin</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Any food intolerance</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Do you have any other health problems besides the above mentioned ones?

☐ No  ☐ Yes (please describe):

_____________________________________________________________________________

Lifestyle and diet:

Are you following a special diet?  ☐ No  ☐ Yes (please describe)

_____________________________________________________________________________

Do you smoke?  ☐ Yes  ☐ No

If yes, how many cigarettes per day?  ☐ < 10 cigarettes/ day  
☐ > 10 cigarettes/ day

If you are a past smoker, how many cigarettes did you smoke per day and when did you quit?

_____________________________________________________________________________

Please list type, duration and frequency of any regular exercise (including walking):

_____________________________________________________________________________

Please indicate the number of alcoholic beverages (spirit 1.5 oz, beer 1 bottle, wine 1 200 ml glass) consumed per day:

☐ < 3/day  ☐ ≥ 3/day

Please indicate the number of coffee drinks per day (1 cup = 1.5 fl.oz.) indicating the type of coffee consumed (filtered, espresso, boiled, etc.)

☐ 0-5 cups/ day  ☐ 6-8 cups/day  ☐ ≥ 9 cups/ day

Type of coffee:________________________________________

Have you participated in a clinical trial within the last month?  ☐ Yes  ☐ No

WOMEN ONLY:

Are you pregnant or breastfeeding?  ☐ Yes  ☐ No

If not, what type of birth control method do you use? (e.g., Barrier or hormonal methods, implantable devices, permanent birth control methods)

Are you post-menopausal?  ☐ Yes  ☐ No

P.I Dr. Vuksan, V. KWG in Type 2 diabetes or Metabolic syndrome  Ver.2 11/29/12
Did you recently experience any of the following symptoms?

<table>
<thead>
<tr>
<th>SYMPTOM</th>
<th>No</th>
<th>Yes</th>
<th>Onset date</th>
<th>Frequency</th>
<th>Duration</th>
<th>Severity (mild/moderate/severe)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bloating</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Belching</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Flatulence (gas)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Diarrhoea</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Excessive urination</td>
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