THE INFLUENCE OF REPORTED PHYSICAL ACTIVITY AND BIOLOGICAL SEX ON CAROTID ARTERIAL DISTENSIBILITY IN CANADIANS WITH DIABETES

THE INFLUENCE OF REPORTED PHYSICAL ACTIVITY AND BIOLOGICAL SEX ON CAROTID ARTERIAL DISTENSIBILITY IN CANADIANS WITH DIABETES

By CONNOR ADRIAN DROOG, Hon.B.Kin

A Thesis Submitted to the School of Graduate Studies in Partial Fulfilment of the Requirements for the Degree Master of Science in Kinesiology

McMaster University MASTER OF SCIENCE (2021) Hamilton, Ontario

TITLE: The influence of reported physical activity and potential sex differences on carotid arterial stiffness in aging Canadians with diabetes

AUTHOR: Connor Adrian Droog, Hon.B.Kin

SUPERVISOR: Dr. Maureen MacDonald

NUMBER OF PAGES: xiv, 97

LAY ABSTRACT

It is well-known that aging is associated with increases in arterial stiffness, which is the progressive impairment of the ability of the arteries to respond to changes in blood pressure and flow. Increased arterial stiffness is associated with the development of cardiovascular disease and appears to be accelerated in females and individuals with diabetes. Physical activity has been highlighted as a potential moderator of age-induced arterial stiffening. Healthy and physically active older adults typically display reduced arterial stiffening versus their more sedentary counterparts, but the extent to which physical activity attenuates vascular stiffening in older adults with diabetes is unclear. Our results, from a cohort of approximately 2000 older Canadians with Type 2 and other diabetes show that self-reported physical activity level does not appear to be associated with arterial stiffness in this population, and these results do not differ between men and women. Follow-up analysis should be conducted to assess the impact of physical activity over time on arterial stiffness in this population.

ABSTRACT

Aging is associated with increases in carotid arterial stiffness, and this process appears to be accelerated in older adults with type 2 diabetes mellitus (T2DM). It is known that older adults with higher levels of physical activity (PA) tend to have lower arterial stiffness values compared to their more sedentary counterparts. Women typically experience a increase in arterial stiffness and cardiovascular events after menopause compared to older men. It is currently unknown whether a greater degree of PA modulates the vascular response with aging in individuals with diabetes and whether sex-based differences exist. We examined arterial stiffness estimated from carotid artery ultrasound images and blood pressure data available from the Canadian Longitudinal Study on Aging (CLSA) baseline data set in participants with Type 2 and other diabetes (DM2O). Arterial stiffness was expressed as carotid artery distensibility, a measure of local arterial stiffness and calculated as the relative change in arterial diameter for a given change in pressure. PA was assessed via the Physical Activity Scale for the Elderly (PASE), a brief 12-item survey used to assess usual PA in adults 65 and older. This study evaluated the association between known cardiovascular disease risk factors/markers and carotid artery distensibility and examined the influence of PA on arterial stiffness. The influence of age and self-reported sex, while controlling for known cardiovascular disease risk factors and markers was examined in individuals with DM2O. There was no effect of PASE score on arterial distensibility before (P = 0.143) and after (P = 0.998) adjusting for known cardiovascular risk factors, and there were no interactions between PASE and sex, or PASE and age. There was a main effect of age on arterial distensibility in both models (P=<0.001), and there was a main effect of sex on arterial distensibility in the final adjusted model only (P=0.040). These findings suggest that PASE is not associated with arterial distensibility in older adults with DM2O, and

these results do not differ by age or sex. Follow-up analysis using longitudinal models is required to further assess the influence of PA on vascular aging.

ACKNOWLEDGEMENTS

First, I would like to thank my supervisor, Dr. Maureen MacDonald. It has been a privilege to be a member of the Vascular Dynamics Laboratory, both as a student and research assistant. Thank you for the opportunities you have provided me that have allowed me to grow, both as a student and a professional. Thank you for the adjustments made at the onset of this pandemic that provided me an uninterrupted transition into research from home and for your support and trust with this project.

Thank you to my committee members, Dr. Baraa Al-Khazraji and Dr. Tom Hawke, for your flexibility and insightful consultation throughout this research process. It has been a pleasure to work with both of you.

Thank you to Dr. Mike Riddell for providing your expertise on this project. Thank you to Dr. Jennifer Voth for your patience, insight, flexibility, and teachings. Your assistance with this work was invaluable.

Thank you to my fellow analyst Joshua Turner for his hard work, support, and Excel wizardry. So much of this possible thanks to his efforts. Thank you to my fellow VDL members, Jenny, Jem, Sydney, Josh, and Emily for creating such a positive and collaborative space. I felt nothing but welcome since arriving at McMaster, and I feel honoured to have worked alongside all of you. I wish nothing but the best for all of you moving forward, and I can't wait to see what amazing things you will all accomplish. Thank you to VDL member Kenny for being a sounding board, numbers guy, and accountability officer, but above all else, a friend. Thank you to all my fellow Kin graduate students at McMaster. Although our time together in person was cut short, the community that exists in this department in unparalleled, and something I will always be proud to have been a part of. I can't wait until the Heimbecker Cup is back on.

Thank you to my partner Carol's family. I am forever grateful for your encouragement, positivity, and patience while I spent countless night couped up in your office, drinking all of your coffee. I can't wait to celebrate with all of you.

Thank you to my loving and supportive parents, without whom I would have never developed the passion and drive that brought me here. Your unconditional love and support were pillars for me through this whole experience, and I am grateful I was able to share all of this with you. Thank you for being the voices of reason and encouragement. To my sister Lauren, thank you for always listening to my scientific rambles and theories. Thank you for being a wonderful and validating listener, and occasional cheerleader, no matter what.

Finally, thank you to my partner Carol. This whole experience brought us closer together than we thought possible, as in, we never thought we could share a desk for this long. This would not have been possible without you. You have been my rock, my sounding board, my biggest supporter, and my most honest editor. Thank you for encouraging me to come to McMaster and to pursue new opportunities, for teaching me the ways of Lot M, and for your unwavering patience, understanding, love, and support. I will cherish our time together at school and cannot wait for what is next. Thank you.

TABLE OF CONTENTS

LIST OF APPENDICES

LIST OF TABLES

LIST OF EQUATIONS

(1) $$\text{Distensibility} = \frac{\Delta\text{Cross- Sectional Area}}{\text{Pulse Pressure} \times \text{Minimum Cross- Sectional Area}}$$

LIST OF ABBREVIATIONS

2hPG in a 75 g OGTT	2-hour (2-h) post-load plasma glucose after a 75 g oral glucose tolerance test
ANG-II	Angiotensin II
baPWV	Brachial-ankle pulse wave velocity
BMI	Body Max Index
BP	Blood pressure
CAC	Central arterial compliance
CAPI	Computer-assisted personal interviewing
CATI	Computer-assisted telephone interview
CCA	Common carotid artery
cfPWV	Carotid-femoral pulse wave velocity
cIMT	Carotid Intima-Media Thickness
CLSA	Canadian Longitudinal Study on Aging
CSA	Cross- sectional area
CTX	c-telopeptide of type 1 collagen
CV	Cardiovascular
CVD	Cardiovascular diseases
DBP	Diastolic blood pressure
DM	Diabetes mellitus
DM2O	Type 2 and other diabetes
DWS	Diastolic wall stress
EGG	Electrocardiogram

eNOS	NO-synthase (eNOS)
FPG	Fasting plasma glucose
HbA_{1c}	Glycated hemoglobin
HDL-C	High-density lipoprotein cholesterol
hsCRP	High-sensitivity C-reactive protein
IDDM	Insulin dependent diabetes mellitus
IEM	Incremental elastic modulus
IL	Interleukin
IMT	Intima-media thickness
LDL-C	Low-density lipoprotein cholesterol
LPA	Light intensity physical activity
LVH	Left ventricular hypertrophy
MVPA	Moderate-vigorous intensity PA
NO	Nitric oxide
PA	Physical activity
PASE	Physical Activity Scale for the Elderly
PIP	Pro-Collagen Type 1 C-Peptide
PP	Pulse Pressure
PWV	Pulse-wave velocity
SBP	Systolic blood pressure
T1DM	Type 1 diabetes mellitus
T2DM	Type 2 diabetes mellitus
TC/HDL Ratio	Total cholesterol/high density lipoprotein cholesterol ratio

VSMC	Vascular smooth muscle cells

CHAPTER 1: LITERATURE REVIEW

1.1 Introduction

Cardiovascular diseases (CVDs) include diseases of the heart and vasculature, such as coronary heart disease, heart failure, peripheral arterial disease, and cerebrovascular disease [1,2]. CVDs are considered one the leading causes of death, accounting for 31% of deaths worldwide [1]. CVD is a major health concern of the aging population. The American Heart Association reported that CVD incidence in the US is ~40% between 40–59 years of age, ~75% between 60–79 years of age, and ~86% in those above the age of 80 [3]. While age itself has been identified as an independent risk factor for CVD, the presence of additional risk factors associated with aging can exacerbate this risk [4]. These include hypertension, obesity, inflammation, and diabetes.

Diabetes mellitus (DM) is a metabolic disorder characterized by elevated blood glucose levels due to the impaired secretion and/or action of insulin [5]. Although an independent risk factor of CVD, diabetes can also be a precursor to hypertension, obesity, and chronic inflammation. Diabetes and aging are both implicated in the progression of arterial stiffness. With aging, arterial walls thicken and lose elasticity, resulting in impaired vascular function, especially in the central arteries located proximally to the heart.[6]. This process is accelerated in the diabetic state [7–11], which highlights the need for therapeutic interventions to address the risk factors of this population.

Physical activity (PA) is a potential mechanism by which individuals can reduce their risk of CVD. Higher PA is associated with lower arterial stiffness in healthy older adults who are more physically active, and those with lower levels of physical activities are associated with higher levels of arterial stiffness [12–15]. Higher levels of PA are also associated with lower concentrations of serum inflammatory markers, highlighting a potential mechanism for PA-

induced improvements in arterial health [16]. A gap in the literature currently exists regarding the influence of PA on arterial stiffness in older adults with diabetes.

1.2 Arterial System

1.2.1 Arterial Anatomy

Arterial walls are composed primarily of elastin and collagen and are divided into three distinct regions: the intima, the media, and the adventitia [17,18]. The intima is the innermost layer, composed of a single layer of vascular endothelial cells with a basement membrane of internal elastic lamina [18,19]. The tunica intima creates the tube-like lumen, through which oxygen-rich blood can be transported to the tissues of the body [18]. The media is the middle layer, and it is comprised of smooth muscle cells and elastin fibres that determine the mechanical properties of the artery, allowing for the regulation of blood flow. The outermost layer of the artery is the adventitia [18] and comprises connective tissue made up of collagen and elastin fibres [19,20]. This connective tissue allows the adventitia to connect the arteries to other tissues in the body, including vascular nerves responsible for smooth muscle cell control [18]. In this way, the structural properties of the artery are responsible for the mechanical functions of the artery.

1.2.2 Arterial Function

The arterial system acts as a conduit to deliver blood at high pressures to the peripheral vascular beds to deliver oxygen, nutrients, and hormones to meet the metabolic requirements of tissues [18,21]. The arterial system can be divided into three distinct regions: the large elastic arteries, the muscular arteries, and the arterioles. The large elastic arteries include the aorta, carotid, and iliac arteries. These arteries contain minimal smooth muscle cells and act as a buffering reservoir to ensure adequate delivery of blood to the capillaries throughout the cardiac cycle. Regulation of delivery is achieved through passive expansion of the artery to allow storage

of the ejected blood during diastole and propulsion of the blood during systole. The muscular arteries (e.g., femoral, popliteal, femoral posterior) serve to modify the travel speed of both pressure and flow waves along their length and determine when the reflected waves will arrive back at the heart in the cardiac cycle by modifying smooth muscle cell tone. Finally, the arterioles can alter peripheral resistance and thus aid in mean arterial blood pressure (BP) maintenance by adjusting their diameter. Chronic changes occur within the central elastic arteries over time, such as progressive carotid arterial stiffening with increasing age, while changes to muscular arteries and arterioles tend to be more acute [21].

1.3 Arterial Stiffness

Arterial stiffness describes the impaired ability of the artery to constrict and dilate in response to pressure changes [22]. Progressive arterial stiffening occurs because of both structural and functional changes to the artery. Vascular alterations are due to hemodynamic forces (pulsatile flow, vascular impedance, wave propagation, and vascular compliance) and extrinsic factors, such as hormones and glucose regulation [23,24]. This progressive arterial stiffening occurs most profoundly in the central and conduit arteries, compared to the more muscular peripheral vasculature. The structural integrity and compliance of the arterial wall are dependent upon the relative contribution of elastin and collagen, the scaffolding proteins. These components are held in balance through cycles of protein production and degradation in response to various stimuli and conditions [24]. Through inflammatory processes, such as those experienced with DM, collagen production can be overstimulated, and elastin production can be impaired, altering the tightly controlled ratio and resulting in stiffer, less elastic arteries [25,26].

1.3.1 Aging

The population of the world is aging. It is expected that the number of adults over 65 will double by the year 2050 [27]. Arterial stiffening is of particular concern, as aging has been highlighted as the main cause of arterial stiffness [28]. Increased arterial stiffness is linked to an increased risk of developing hypertension and atherosclerosis, both of which can result in CVD and mortality [17,22]. Arterial stiffness is also associated with left ventricular hypertrophy (LVH) and coronary artery disease [29]. Indeed, Framingham Heart Study findings support the notion that increased arterial stiffness is associated with an increased risk of a cardiovascular (CV) event [30]. Potential mediators of this age-induced vascular stiffening, thus, require further investigation.

Aging is associated with an increased risk of arterial dysfunction due to two distinct processes: structural changes in the vascular walls (structural) and reductions in endothelial function (functional) [31]. It is well-established that aging is associated with the progression of carotid artery stiffening. Over time, arterial walls thicken and lose elasticity, resulting in a reduction in the buffering function of the central conduit arteries [6]. This arterial stiffening is due, in part, to the fragmentation and loss of elastin fibres and the aggregation of stiff collagen fibres in the walls of the vasculature [17]. As such, with aging large elastic arteries display a reduction in their low-stretch bearing component and shift the load to the stiffer matrix components (collagen) [32]. This shift impairs the buffering function of the large arteries, the purpose of which is to ensure continual anterograde blood flow during diastole [33]. As a result of this reduced elasticity and impaired buffering function, more blood must be transported over longer distances following systole, requiring higher systolic pressures and cardiac strain. These combined changes throughout the cardiac cycle increase the diastolic-systolic difference (pule pressure) also result in a greater mechanical strain on the downstream vessels, organs, and tissues [32]. These

vascular changes are due to progressively increasing oxidative stress and chronic low-grade inflammation, which appear to function synergistically in the progression of arterial stiffness [24,34].

The vascular endothelium lines the internal lumen of all blood vessels and provides an interface between the blood and the vascular smooth muscle cells (VSMC) [35,36]. The endothelium is also a regulator of the VSMC, via several chemical mediators that contribute to the maintenance of vascular homeostasis [35,36]. Central to this regulatory process is nitric oxide (NO), produced by the enzyme endothelial NO-synthase (eNOS) [37]. eNOS is responsible for generating NO linked to vascular relaxation (vasodilation) and responds to both chemical and mechanical stimuli [36]. In response to these stimuli, NO is generated and can diffuse to the VSMC, resulting in dilation of the artery [36]. This control of vasomotor tone is imperative for the balance of tissue oxygen and metabolite delivery [35]. Endothelial cell function begins to decline in early middle age [38]. The decline in endothelial cell function is thought to be due to age-induced increases in ROS, likely from dysfunctional mitochondrial cells and oxidant enzymes such as NADPH oxidase [39]. ROS scavenge available NO, thus decreasing the bioavailability of NO. Chronic low-grade inflammation due to increases in circulating inflammatory mediators works synergistically with circulating ROS, impairing endothelial function and increasing arterial stiffness [39]. As such, progressive increases in chronic low-grade inflammation and circulating ROS are associated with structural and functional changes to the artery, resulting in stiffer arteries and a greater risk of a CV event [39].

1.3.2 Carotid Intima-Media Thickness

Carotid intima-media thickness (cIMT) is a measure of arterial wall thickness, and like arterial stiffness, increases with age [40]. cIMT can be used to measure the progress of

atherosclerosis and is associated with CV risk factors in older healthy and diabetic populations [40]. cIMT can be assessed noninvasively *via* ultrasound imaging of the carotid artery [41]. Arterial stiffness and cIMT reflect independent indices of vascular damage, however, thickening of the intimal wall of the carotid artery appears to be due primarily to increases in carotid systolic blood pressure that occur with aging [42]. Both carotid arterial stiffness and thickness appear to be independent predictors of CV risk in aging adults and are processes that can be exacerbated in certain clinical populations [43].

1.3.3 Sex differences

It is well-established that arterial stiffness and risk of CV events both increase drastically in females following menopause than males of the same age [44]. Given the cardio-protective effects of estrogen, it is proposed that the substantial increases in arterial stiffness following menopause in females is due to reductions in estrogen [19]. In a study of 3149 females between the ages of 21-90, it was found that post-menopausal females displayed the highest rates of arterial stiffness as assessed by brachial-ankle pulse-wave velocity (baPWV) and carotid artery compliance [45]. Infusion of ascorbic acid resulted in improved function in late and post-menopause only, suggesting the potential role of oxidative stress in the accelerated vascular stiffening following menopause [46]. It has been found that central arterial stiffness assessed via carotid-femoral PWV is significantly reduced following 4 months of estrogen treatment (0.625 mg/d of conjugated equine estrogen) in postmenopausal females [47]. This highlights not only the cardioprotective effects of estrogen, the loss of which may be responsible for the marked difference in CVD risk in older females compared to older males, but the recoverability of arterial function in this population.

Sex differences exist in the age-associated increases in cIMT as well, such that certain CV risk factors exert differential effects based on sex [48,49]. For example, one study found that cIMT values were significantly lower in females versus males over the age of 45, and that sex differences exist in the predictive value of certain CV risk factors for determining cIMT [49]. In that same study, only age, hypertension, and Type 2 diabetes mellitus (T2DM) were predictive of cIMT in males, but age, pulse pressure, metabolic syndrome, and waist circumference were predictive of cIMT in females [49]. The exact mechanism regarding this reported differential impact of CV risk factors in males versus females remains unknown, however, the impact of metabolic syndrome is reportedly more pronounced in females due to premature arrest of the protective effects of estrogen on the CV system [49].

1.3.4 Assessment of Arterial Stiffness

Pulse-wave velocity, specifically in the carotid-femoral region, is considered the gold standard metric of assessing arterial stiffness in humans [50]. PWV is a direct, non-invasive, simple, and reproducible method for assessing arterial stiffness. The carotid-femoral region specifically refers to the aorta-iliac pathway is considered the most physiologically relevant, as the aorta and its branches are directly connected to the left ventricle and thus strongly implicated in the pathophysiology of arterial stiffening. PWV is most commonly assessed using the foot-to-foot velocity method applied to pressure waveforms. The waveforms are typically obtained at the right common carotid artery and right femoral artery, and the transit time is assessed between the feet of the waveforms. At the end of the diastolic phase, the foot of the wave is noted at the onset of the steep rise in the waveform. The transit time is the time of travel of the foot of the wave over a measured distance. A faster transit time is indicative of more rapid waves and thus greater arterial stiffness. Assessment of PWV can be difficult to conduct in populations with diabetes or

obesity, as the transcutaneous assessment of the femoral waveform can be difficult to capture, and the distance measurement may be inaccurate [50].

Arterial compliance is the absolute diameter change for a given pressure at a controlled length and is indicative of overall arterial health and the buffering capacity of the heart [51,52]. Arterial distensibility is a measure of the relative change in diameter for a given change in pressure [51]. Distensibility is a measure of local arterial stiffness in large elastic arteries and is primarily a determinant of arterial wall stress [29,51]. As such, arterial compliance and distensibility are typically related to each other, as compliance can be calculated using the distensibility and arterial volume (*Compliance = Distensibility x Arterial volume*) [52]. However, compliance and distensibility can reflect different health indices, as compliance has been shown to be an independent predictor of both CVD and death, while distensibility is an independent correlate of mortality only [29]. A decrease in arterial distensibility, and thus the ability of the artery to respond to changes in pressure, reflects an increase in arterial stiffness. Functional impairment to the vascular wall may become apparent prior to any observable and attributed structural changes attributed to the atherosclerotic process and before any overt symptoms of CVD [53].

Distensibility of the common carotid artery (CCA) can be assessed directly and non-invasively using B-mode ultrasound, which allows for simultaneous image acquisition of arterial wall thickness and vessel diameter throughout the cardiac cycle [54]. The change in arterial diameter is dependent upon the change in arterial pressure. Calculation of arterial distensibility also requires measurement of blood pressure [32]. Blood pressure can be assessed locally via applanation tonometry on the carotid artery opposite to the one being imaged, or peripherally, using a standard brachial blood pressure cuff [32,54]. Local BP values can differ from brachial BP values due to pulse amplification as it travels down the vessel [32]. As such, local assessment of BP

is typically recommended; however, this may be limited by equipment availability, and tonometry can be difficult to assess in certain clinical populations, such as DM [32,55]. Distensibility can be calculated using the following equation:

$$(1)\ \text{Distensibility} = \frac{\Delta\text{Cross- Sectional Area}}{\text{Pulse Pressure} \times \text{Minimum Cross- Sectional Area}}$$

Wherein Δcross-sectional area (CSA) is the difference between the maximum and minimum CSA, and pulse pressure is the difference between resting systolic and diastolic blood pressure.

1.4 Diabetes Mellitus

DM is a metabolic disorder characterized by chronic hyperglycemia due to the impaired secretion and/or action of insulin [5]. This chronic hyperglycemia is associated with microvascular impairments and an increased risk of CVD. Diagnostic criteria for diabetes exist, and they are based on glycemic thresholds associated with microvascular disease [5].

There are currently four recommended diagnostic tests for diabetes. These include the measurement of fasting plasma glucose (FPG); 2-hour (2-h) post-load plasma glucose after a 75 g oral glucose tolerance test (2hPG in a 75 g OGTT); glycated hemoglobin (HbA_{1c}); and a random blood glucose test when other signs and symptoms of diabetes are present [56] . $HbA_{1c,}$ obtained from the blood, is a gold standard metric of long-term glycemic control [57]. People with FPG values of ≥ 7.0 mmol/L (126 mg/dl), 2-h post-load plasma glucose (2hPG in a 75 g OGTT) ≥ 11.1 mmol/L (200 mg/dl) [58], HbA_{1c} ≥ 6.5% (48 mmol/mol); or a random blood glucose ≥ 11.1 mmol/L (200 mg/ dl) in the presence of signs and symptoms are considered diabetic [59]. Individuals with an FPG of 6.1-6.9 mmol/L are considered to have impaired fasting glucose. Individuals with a 2hPG in a 75 g OGTT value of 7.8-11.0 mmol/L are considered to have impaired glucose tolerance. Individuals with an HbA_{1c} value between 6.0-6.4% are considered prediabetic. Complications can still arise from prediabetes, as these individuals are at risk for

developing diabetes and all associated complications [5] . When using HbA_{1c} to diagnose diabetes, it is important to recognize that HbA_{1c} is an indirect measure of average blood glucose levels, and other factors should be considered that may impact hemoglobin glycation independently of glycemia, including age, race/ ethnicity, pregnancy status, genetic background, and anemia/hemoglobinopathies. Diabetes typically manifests as either type 1 or 2 diabetes mellitus. Less common forms such as gestational, which occurs during pregnancy, or monogenic diabetes, which results from a mutation of a single gene as opposed to the polygenic type 1 and 2 [59–61].

1.4.1 Type 1 Diabetes Mellitus

Type 1 diabetes mellitus (T1DM), also known as insulin-dependent diabetes mellitus (IDDM), is a chronic autoimmune disorder characterized by the selective destruction of insulin-producing pancreatic beta cells [62,63]. In healthy populations, blood glucose levels increase immediately following a meal, stimulating insulin production [64]. However, individuals with T1DM require exogenous insulin, in the form of insulin bolus injections or a continuous basal dose through an insulin pump with additional administration around meals, to maintain normal blood glucose levels [62]. Insulin is also a vasoactive hormone that acts on larger conduit arteries at the macro-vascular level to increase vascular compliance and acts on resistance arterioles and precapillary arterioles at the microvascular level, to increase vascular flow and perfusion of capillaries through vasodilation, respectively [65].

Arterial stiffening has been noted in individuals with T1DM of varying ages [7,8]. The structural and functional characteristics of the CCA were assessed in 45 children and adolescents with T1DM between the ages of 9-19 years old, with no reported microalbuminuria, elevated lipids, or hypertension. Individuals were age, sex, and body mass index (BMI)-matched with controls, differing only in diabetic status. Intima-media thickness (IMT), compliance,

distensibility, diastolic wall stress (DWS) and incremental elastic modulus (IEM) of the CCA were assessed. DWS provides a metric of the stress on the arterial wall during diastole and is associated with left ventricular hypertrophy [66]. IEM reflects the ability of the wall to resist elastic deformation, or properties of the arterial wall independent of the arterial geometry [67]. Compared to controls, children with T1DM had significantly greater vales for IMT, DWS, and IEM. However, no significant differences were noted for compliance or distensibility. Multivariate regression analysis revealed that a diabetic state was the strongest predictor of IMT, DWS, and IEM, and these results were interpreted as indicative of early structural impairment in children with T1DM [7].

Structural alterations may precede impairments in carotid arterial distensibility Giannattasio *et al.* (1999) assessed the distensibility of the CCA in 133 adults (74 males, aged 35 ± 0.9 years) with T1DM and no macrovascular complications. Arterial distensibility was assessed via B-mode ultrasonography of the CCA, and blood pressure was assessed at the brachial artery. It was found that, relative to 70 age-matched normotensive controls, arterial distensibility was significantly reduced in adults with T1DM by an average of 14%. Similar to the previous findings in children, cIMT was also significantly greater in the T1DM group [7]. Carotid artery distensibility was found to be inversely related to age, duration of disease, and systolic blood pressure, all of which were significantly associated with cIMT. As such, it does appear that T1DM is associated with increased cIMT early in life and decreases in arterial distensibility in young-middle-aged adults with T1DM. These findings are indicative of early vascular stiffening in this population.

Similar results regarding accelerated vascular stiffening in adults with T1DM have been corroborated using other indices of vascular stiffness as well. Llauradó *et al.* (2012) assessed

arterial stiffness via aortic PWV and the serum inflammatory marker high-sensitivity C-reactive protein (hsCRP) in adults with T1DM and no CVD (n= 68; 34 males, 34 females; 35 ± 9 years) and 68 healthy age and sex-matched controls. Individuals with T1DM displayed greater PWV, indicating increased stiffness, compared to healthy controls, even after adjusting for other cardiovascular risk factors. Males had higher concentrations of hsCRP, and aortic PWV and hsCRP were positively correlated in males only. This study highlights chronic inflammation as a potential mechanism by which a diabetic state can induce accelerated arterial stiffening and suggests sex-based differences in this mechanism [9]. In summary, arterial stiffness as a CV risk factor in T1DM, as it has been shown that arterial stiffness is predictive of mortality in this population, independent of other CV risk factors [68].

Potential sex differences in the degree of impairment to arterial distensibility have been noted in T1DM as well. Rydén Ahlgren *et al.* (1995) assessed distensibility of the CCA in 30 females (aged 20-61 years) and 26 males (aged 22-56 years) with T1DM and healthy age and sex-matched controls. Females with T1DM displayed significantly greater arterial stiffness compared to healthy females, but no difference was found in males [10]. More recent data have since demonstrated accelerated stiffness in both males and females with T1DM [9]. This contrasts with the findings from Rydén Ahlgren *et al.* (1995) who included participants with a range of ages, which may have confounded the data by failing to account for the vascular implications of menopause.

1.4.2 Type 2 Diabetes Mellitus

Prediabetes and T2DM are often indicative of an underlying disorder, such as the metabolic syndrome [69]. This highly prevalent condition is characterized by health complications such as abdominal obesity, hypertension, dyslipidemia and hyperglycemia [5]. The prevalence of

T2DM accounts for 90-95% of diabetes, and β-cell dysfunction is required to develop T2DM. Most individuals with T2DM have relative insulin deficiency, but insulin levels have been observed to increase early in the course of the disease due to insulin resistance [60]. In contrast with T1DM, most people with T2DM are either overweight or obese, which either causes or can exacerbate, their resistance to insulin [5,70,71]. Many individuals who are overweight but not obese by BMI criteria (>30 kg/m^2) have a higher proportion of primarily abdominal body fat, which is indicative of greater visceral adiposity compared to people without diabetes [72]. Insulin treatment may not be required for survival but may be required for glycemic control purposes [73].

It is well-accepted that arterial stiffening and thickening are exacerbated in individuals with T2DM, and these events primarily occur at the central rather than peripheral arteries [11,74]. Charvat *et al.* (2010) assessed the distensibility and thickness of the CCA in 82 older, normotensive adults with diabetes (54 males, 28 females; aged 61 ± 6 years) and 41 age-matched controls. Distensibility and cIMT were assessed via B-mode ultrasonography at the CCA. Distensibility was significantly reduced in individuals with T2DM compared to controls (0.27 ± 0.11 vs. 0.37 ± 0.16), but no significant differences in cIMT were noted. It was found that distensibility was significantly associated with both disease duration and BMI in individuals with T2DM [75]. These findings of no difference in cIMT between groups are in contrast with those from Al-Nimer & Hussein (2009), who assessed the cIMT of 46 (45-77 years old) adults with T2DM with, and without, normotensive metabolic syndrome. The individuals with normotensive metabolic syndrome displayed significantly greater cIMT values than those with metabolic syndrome (0.824 ± 0.155 vs. 0.708 ± 0.113, respectively) [75]. It is possible that while both Charvat *et al.* (2010) and Al-Nimer & Hussein (2009) assessed normotensive individuals with T2DM, the T2DM group from Charvat *et al.* (2010) might not have all displayed the metabolic

syndrome; however, the necessary classification criteria [76] for this was not reported by this group.

Accelerated vascular stiffening in T2DM has also been corroborated using other indices of stiffness. Cameron *et al.* (2003) assessed arterial stiffness across three arterial segments via PWV and central arterial compliance (CAC) in a large cohort of healthy middle to older-aged adults (age 34-90) (n=169; 100 males [39 T2DM]; 69 females [18 T2DM]. In this study, PWV was assessed centrally along the carotid-femoral region and peripherally along the carotid-radial region and from the aorta to the finger. Despite being an average of 10 years younger, individuals with T2DM displayed lower CAC and faster PWV (both indicating increased arterial stiffness) at all measurement sites in comparison to their non-diabetic counterparts [74]. CAC is primarily indicative of the buffering capacity of the aorta, and it appears to be impaired to a greater extent, and at an earlier age, in individuals with T2DM. This study highlights the accelerated rate at which adults with T2DM experience impaired vascular function (decreased arterial compliance and increased arterial stiffness) compared to their non-diabetic counterparts.

Accelerated and preferential impairment of the central, but not peripheral, arterial segments in individuals with T2DM was further corroborated by Kimoto *et al.* (2003). Central (heart-carotid and heart-femoral) and peripheral (heart-brachial and femoral-ankle) PWV were assessed in 161 older adults with T2DM (85 males and 76 females; mean age of 60 years) and 129 healthy control participants (56 males and 73 females; mean age of 59 years). Individuals with T2DM had elevated PWV in all regions assessed, however, PWV was elevated more in the central arterial regions. Furthermore, the effect of age on arterial stiffness was also largest at the central rather than peripheral regions of assessment, such that PWV increased more in older individuals (with and without T2DM) at the central regions. This study further highlights the

localization of increases in arterial stiffness with aging, and the acceleration of this process in individuals with T2DM [11]. A similar study from Gómez-Marcos et al. (2011) further corroborated these findings and additionally found that cIMT, assessed via ultrasound imaging, was thicker in middle-older aged adults (aged 44-66) with T2DM.

1.4.3 Diabetes, Inflammation, and Accelerated Vascular Aging

This accelerated vascular aging in a diabetic state appears to be related, and due in part, to chronic low-grade inflammation [77–79]. It is known that individuals with poorer glycemic control display elevated levels of chronic inflammation. In a large sample of adults with diabetes (n = 1614; age 17-66+), it was found that increased HbA_{1c} was associated with higher levels of hsCRP, and thus chronic inflammation [79]. This relationship translates to accelerated vascular stiffness in diabetic populations [78]. In a group of 362 middle-aged and elderly males (mean age 60 ± 11 years), Nakhai-Pour et al. (2007) assessed aortic PWV and hsCRP. It was found that individuals with diabetes mellitus who were older had higher levels of hsCRP. Furthermore, it was found that hsCRP was predictive of PWV and thus arterial stiffness, such that aortic PWV increased significantly with higher levels of hsCRP [78]. Arterial stiffness is also associated with increased activity of angiotensin II (ANG-II), a known vasoconstrictor [77,80]. The activity of ANG-II results in the downstream activation of increased production of reactive oxygen species and inflammatory cytokines, which result in the stimulation of C-reactive protein (CRP) production in the vascular smooth muscle cells. CRP promotes vascular inflammation and endothelial dysfunction, both of which further increase vascular stiffening [77]. A diabetic state results in elevated levels of hsCRP, a marker of chronic inflammation, which further exacerbates the age-related process of vascular stiffening in individuals with DM.

1.5 Known Risk Factors & Markers of Arterial Stiffness

1.5.1 HBA_{1c}

HbA_{1c} reflects long-term glycemic control, typically reflective of the status in the 8-12 weeks before sample acquisition. HbA_{1c} is reported as a percentage of total hemoglobin, and it is assessed via blood draw. Values are standardized using the National Glycohemoglobin Standardization Program (NGSP) to compare testing sites [81]. Meta-analyses have confirmed that increases in HbA_{1c} are associated with increases in CV risk, such that for every 1% increase in HbA_{1c}, the relative risk for any cardiovascular disease event is 1.17-1.18 for individuals with T2DM and 1.15 for individuals with T1DM [82,83]. As such, chronic hyperglycemia and thus elevated HbA_{1c} is associated with increased risk of CVD and all-cause mortality in adults with DM [83]. Both chronic hyperglycemia and fluctuations in glycemic control can initiate inflammatory responses through increased stress on the endoplasmic reticulum and mitochondrial superoxide production. Chronic inflammation and oxidative stress due to hyperglycemia are implicated in the pathogensis of endothelial dysfunction, which can be followed by atherogenesis and an increased probability of cardiovascular event. Glucose toxicity and the associated oxidative stress and inflammation act in a cyclical fashion that results in impaired insulin sensitivity, the loss of β-cells, and vascular dysfunction [84]. HbA_{1c} is an independent predictor of arterial stiffness and endothelial dysfunction in adults with T2DM, especially when complicated by hypertension [85].

1.5.2 C-Reactive Protein

CRP is an inflammatory risk marker and is involved in atherosclerosis. As a risk marker, it is expressed as hsCRP and is measured via blood draw and either immunonephelometric or immunoturbidimetric assay. It is primarily synthesized and secreted by

hepatocytes in response to interleukin (IL)-6 and IL-1, tumor necrosis factor-alpha, or other proinflammatory cytokines. CRP increases the phagocytic activity of neutrophils and induces the expression of adhesion molecules. CRP is involved in the synthesis of tissue factors, cytokines, and platelet aggregation at the site of vascular damage, and as such, it is heavily involved in the atherosclerotic development process [86]. CRP is present in the atherosclerotic lesion at the level of the intima, where it can co-localize with monocytes, monocyte-derived macrophages and lipoproteins. The process of co-localization directly adds to the atherosclerotic development process. Elevated inflammation is implicated in the initiation, development, and destabilization and rupture of atherosclerotic plaque. Endothelial damage by atherosclerotic development leads to a reduction in the bioavailability of NOS and associated decreases in the release and activity of NO, and increases in the degradation of NO, which results in the excess generation of ROS. As such, the endothelial damage caused by the development of atherosclerotic plaque both impairs vascular function and contributes to a proinflammatory environment [87]. CRP is elevated in various conditions, including acute and chronic inflammation, peripheral vascular diseases, diabetes, and hypertension [86]. It has been demonstrated that CRP is associated with increased arterial stiffness assessed via PWV but not distensibility in middle- and older-aged adults [88,89]. Research regarding the distensibility of the CCA and hsCRP is limited, especially in older adults with diabetes, and further investigation is required in this area.

1.5.3 Triglycerides

Dyslipidemia is a risk factor for CVD, with elevations in triglyceride concentrations (>1.7mmol/L) associated with an increased risk of developing CVD [90]. In a large cohort study of males and females between the ages of 20-93 (n = 7587), elevated nonfasting triglyceride levels were associated with a greater risk of myocardial infarction, ischemic heart disease, and

mortality. Elevated triglyceride levels, or hypertriglyceridemia, is a marker for several forms of atherogenic lipoproteins [91]. These include low high-density lipoprotein cholesterol (HDL-C), small low-density lipoprotein (LDL) and high-density lipoprotein (HDL) particles, partially metablozied chlyomicrons (CM), and very low density lipoproteins (VLDL). Independent of LDL, VLDL and CM remnants are atherogenic, as they are prone to accumulation at the endothelium. These remnants can be taken up by macrophages and form foam cells, which promote the formation of fatty streaks in the walls of vasculature, and precede the development of atherosclerotic plaque [91]. Reductions in triglycerides have been noted as a potential therapeutic target for arterial health, as lower levels of triglycerides were associated with lower central arterial stiffness assessed via carotid-femoral PWV [92]. Limited research exists regarding elevated triglycerides and arterial distensibility, especially in individuals with DM. However, lowering triglycerides may reduce residual CV risk, which may be particularly important for individuals with diabetes and dyslipidemia [93].

1.5.4 Cholesterol

Both low-density lipoprotein cholesterol (LDL-C) and high-density lipoprotein cholesterol (HDL-C) are implicated in arterial stiffness and CVD risk [94,95]. LDL-C is the total amount of cholesterol contained within LDL particles, and it is particularly implicated in the atherosclerotic process [96]. Small, dense LDL particles are more likely to be oxidized and taken up by macrophages in the arterial wall, exacerbating the atherosclerotic process. HDL-C opposes the atherosclerotic process by removing cholesterol from foam cells, inhibiting the oxidation of LDL-C, and pro-inflammatory processes [97]. The ratio of LDL-C/HDL-C has been demonstrated to be predictive of CV risk [98]. However, it has been shown in middle-aged males that variations in the ratio of total cholesterol/HDL-C were more strongly associated with risk of ischemic heart

disease and insulin resistance than LDL-C/HDL-C. It was proposed that this observation is due to the limited variation is found in the plasma LDL-C levels in overweight, hyperinsulinemic men [94]. Both are independently associated with central arterial stiffness, such that LDL-C was independently associated with central arterial stiffness, and HDL-C was independently and inversely associated with central and peripheral stiffness in a large cohort of middle-older aged adults (n = 2375, 40-96 years old) [95]. Lower HDL-C is predictive of arterial stiffness in adults with T1DM [99]. Furthermore, non HDL-C cholesterol has been identified as an independent risk factor for CVD in the general population and T2DM [100], and lowering LDL-C and reducing the risk of CV events in T2DM [101].

1.5.5 Blood Pressure

Hypertension, defined as a systolic blood pressure of ≥130 mmHg and diastolic blood pressure (DBP) of ≥80 mmHg, is a highly prevalent contributor to CVD and arterial stiffness [102,103]. Hypertension is highly prevalent in T1DM and T2DM , with an incidence rate of approximately 68% in the United States[104]. Clinically blood pressure is typically assessed with an automated blood pressure device using oscillometry [105]. Oscillometry is an automated system that senses the pressure pulsations within an inflatable cuff wrapped around the bicep or wrist. The cuff is inflated to a suprasystolic pressure, then slowly deflated to a subdiastolic pressure while oscillations in pressure are recorded [105].

Elevated systolic blood pressure is associated with an increased risk of CVD independent of DBP [106]. As a result, SBP is the principal therapeutic target in hypertension cases [107]. SBP tends to increase from age 30 onwards, until at least age 84 [108]. However, in a large cohort of roughly 170 000 individuals from the UK Biobank, while both SBP and DBP were associated with arterial stiffness, midlife DBP was the strongest predictor of an increase in arterial stiffness

over time [109]. Furthermore, arterial stiffness was associated with an earlier transition to reductions in DBP and an earlier risk of developing hypertension or elevated arterial stiffness. DBP tends to increase until the age of approximately 50 years old, then begins to decrease around age 60 until at least the age of 84. This, coupled with the known progressive increase in SBP with age, results in widening pulse pressure (PP) with age [108], thus highlighting the importance of DBP control in midlife to prevent accelerated arterial stiffening [109].

Hypertension is associated with endothelial dysfunction, and oxidative stress has been shown to be a consequence of hypertension [110]. Elevated intraluminal pressures have been associated with reductions in endothelial-dependent vasodilation, increases in the production of superoxides, and increased activity of nicotinamide adenine dinucleotide phosphate (NADPH) oxidase [111]. Furthermore, a hypertensive state has been shown to result in the expression of ROS from NADPH and mitochondria [111]. Excessive ROS production results in a reduction in the bioavailability of NO, and thus endothelial dysfunction [112].

1.5.6 BMI

BMI is calculated by dividing an individual's weight in kilograms (kg) by their height in metres squared [113]. A BMI less than 18.5 kg/m^2 is considered underweight. A BMI between 18.5 to <25 kg/m^2, is considered a healthy weight. A BMI between 25.0 to <30 kg/m^2 is considered overweight. A BMI of 30.0 kg/m^2 or higher is considered obese [113]. Most people with T2DM are either overweight or obese, which either causes or can exacerbate the resistance to insulin [5,70,71]. Many individuals with T2DM who are not obese by BMI criteria have a higher proportion of primarily abdominal body fat, which indicates greater visceral adiposity than people without diabetes [72]. Obesity is associated with a significant increased risk of cardiovascular morbidity and mortality compared to a healthy BMI [114]. Furthermore, being overweight is associated with

a significantly increased risk of developing CVD at an earlier age, and thus a longer exposure time to elevated CVD risk, despite similar longevity of those with a healthy BMI [114]. Concerning arterial stiffness, visceral adiposity is associated with increased arterial stiffness in adults [115]. Excessive visceral adiposity results in the synthesis of excessive adipokines and pro-inflammatory cytokines and thus the chonric inflammation typically exhibited by those with obesity. Systemic inflammation is heavily implicated in the pathogenesis of atherosclerosis and thus the occurance of CV events via increased macrophage infiltration and reductions in the stability of arterial plaques [116].

1.6 Physical Activity & Arterial Stiffness

Individuals who are more physically active and/or those with a higher degree of physical fitness may mitigate age-induced arterial stiffening [12,13,117–121]. Arterial stiffness was assessed via aPWV in 53 healthy pre and post-menopausal females, further stratified by PA level into "sedentary" (n= 28) and "physically active" (n= 25). It was found that age-related increases in central arterial stiffness were present in sedentary females and that this response was attenuated in physically active females. Furthermore, aerobic fitness was a significant independent predictor of central arterial stiffness in healthy older females [117]. Similar results were corroborated by Kozakova *et al.* (2007) in a cohort of 432 healthy middle- and older-age males (n= 166) and females (n= 266) between the ages of 30 and 60. Carotid artery stiffness and cIMT were assessed via B-mode ultrasonography, and PA was assessed objectively via accelerometry. Both carotid arterial stiffness and thickness increased with age in both males and females, and the magnitude of PA was negatively related to arterial stiffness only. Following multivariate regression analysis, age and PA were independently related to carotid arterial stiffness but not thickness. As

such, it appears that habitual PA may attenuate age-related increases in arterial stiffness but not thickness [122].

Results from larger cohort studies are not as conclusive concerning the degree to which PA is associated with arterial stiffness, and these outcomes may be influenced by how PA is assessed. The Atherosclerosis Risk in Communities (ARIC) (1987-1989) study assessed the distensibility of the left CCA using B-mode ultrasonography in 10,644 males and females aged 45–64 years without CVD. PA was assessed using a modified version of the Baecke PA questionnaire [123]. This questionnaire assessed PA from work, leisure, and sport, on a scale of 1 (low) to 5 (high). Participants in this cohort were largely sedentary, and the proportion of participants participating in regular PA was 4.8%. It was found that work activity was weakly associated with arterial distensibility while there were no associations with leisure or sports activity [118]. These findings are in contrast with previous work [12,13,117] and suggest that habitual PA may not be associated with arterial distensibility in middle-older aged healthy adults.

Tanaka *et al.* (2018) outlined several limitations in work from Schmitz *et al.* (2002), including the relatively limited and young age range of the participants (45-64), which may have resulted in a narrow range of arterial stiffness measures, and the use of a single metric of PA in a mostly sedentary group. A subset of 3893 older adults (aged 66-90) from the ongoing ARIC study (2011-2013) were examined prospectively using the same Baecke PA questionnaire. PA data was taken from the first (1987-1989) and third (1993-1995) visits to examine the persistence of PA. Central arterial stiffness was expressed via carotid-femoral PWV in this case, and it was found that higher PA in later life and habitual PA from mid to later life were associated with lower central arterial stiffness. Despite minor attenuation, these findings persisted after adjusting

for diabetes and hypertension [12]. It appears that arterial stiffness is influenced by habitual PA in older adults, and results may differ based on the methodological approach.

Self-reported PA levels are moderately associated with elevated cardiovascular fitness [119]. Older individuals with a higher level of cardiorespiratory fitness may display attenuated arterial stiffness and thickness [120,121]. Moreau *et al.* (2006) assessed intima media thickness of the femoral artery in 173 sedentary, moderately active, and endurance-trained males further stratified by age into young (20-39 years), middle-aged (40-59 years) and older (60-79 years) and 74 pre or post-menopausal females, further stratified by sedentary and endurance-trained. In both males and females, femoral artery IMT increased with age; however, absolute IMT values and the percent change in IMT were lower in endurance-trained compared to sedentary counterparts. While the Moreau *et al*. study assessed changes in femoral and not carotid IMT, similar findings have been reported when assessing cIMT [120,121]. Gando *et al.* (2011) assessed cardiorespiratory fitness via VO_{2peak} in 771 adults (180 males and 591 females) between 27 and 65 years of age. Arterial thickness and lumen diameter were assessed at the CCA. In older individuals (60+), carotid arterial thickness and lumen diameter were lower in the fit group compared to the unfit group. It appears that habitual PA resulting in improved fitness is necessary to attenuate the age-related increases in both arterial stiffness and thickness.

1.6.1 Mechanism of Exercise-Induced Improvements in Arterial Stiffness

The previously observed attenuations in age-related impairments in vascular function and structure may be due, in part, to exercise-induced reductions in hsCRP. One meta-analysis reported that an inverse relationship exists between regular PA and serum hsCRP, such that those who are more physically active display reduced serum markers of hsCRP. The likely mechanism through which PA improves serum inflammatory markers directly via downregulation of pro-

inflammatory cytokine production in adipose and muscle tissue and upregulation of atheroprotective cytokine signalling, resulting in a cardio-protective effect [16].

Progressive exercise-induced alterations in the ratio of structural proteins in the vascular wall are implicated in the cardio-protective effects. It has been demonstrated in overweight, pre-menopausal females that 16-weeks of combined aerobic and resistance training and a hypo-caloric diet resulted in increased levels of c-telopeptide of type 1 collagen (CTX), a marker of type 1 collagen degradation [124]. Increased levels of CTX were found despite unchanged levels of Pro-Collagen Type 1 C-Peptide (PIP), a marker of type 1 collagen synthesis [124]. Increased carotid artery distensibility has since been associated with increases in both CTX and PIP [125].

1.7 Purpose and Hypothesis

What is currently unknown is whether reported PA is associated with accelerated arterial stiffness in aging Canadians with DM2O and whether differences between sexes exist in this population. It is known that age-related arterial stiffening is increased in individuals with T2DM, and it is proposed that it is primarily due to elevations in serum inflammation as indicated by markers such as hsCRP [77–79]. Furthermore, regular PA that results in a higher level of fitness results in attenuated age-induced increases in arterial stiffness, partially through downregulation of hsCRP concentrations [16]. As such, older individuals with T2DM who are regularly physically active may display attenuations in the accelerated vascular aging associated with the condition. Furthermore, it is known that females display an increase in arterial stiffness following menopause compared to males of the same age [44]. The results of this study will also investigate whether sex modulates the relationship between PA and arterial aging.

The specific objective of this thesis is to evaluate the influence of PA and sex on carotid artery stiffness measured in the baseline data sets from a large cohort of older Canadians with DM2O while controlling for known CVD risk factors and markers.

It is hypothesized that like healthy older adults, more physically active individuals with DM2O will have attenuated common carotid artery stiffness in comparison to their more sedentary counterparts. Furthermore, it is hypothesized that sex differences will exist in this relationship, such that accelerated arterial stiffness will be more pronounced in older females than males.

References

1. World Health Organization. *Cardiovascular Diseases (CVDs)*.; 2017.
2. Virani SS, Alonso A, Aparicio HJ, et al. Heart Disease and Stroke Statistics-2021 Update: A Report From the American Heart Association. *Circulation*. 2021;143(8). doi:10.1161/CIR.0000000000000950
3. Yazdanyar A, Newman AB. The Burden of Cardiovascular Disease in the Elderly: Morbidity, Mortality, and Costs. *Clin Geriatr Med*. 2009;25(4). doi:10.1016/j.cger.2009.07.007
4. Rodgers JL, Jones J, Bolleddu SI, et al. Cardiovascular Risks Associated with Gender and Aging. *J Cardiovasc Dev Dis*. 2019;6(2). doi:10.3390/jcdd6020019
5. Punthakee Z, Goldenberg R, Katz P. Definition, Classification and Diagnosis of Diabetes, Prediabetes and Metabolic Syndrome. *Can J Diabetes*. 2018;42:S10-S15. doi:10.1016/j.jcjd.2017.10.003
6. Sun Z. Aging, arterial stiffness, and hypertension. *Hypertension*. 2015;65(2):252-256. doi:10.1161/HYPERTENSIONAHA.114.03617
7. Atabek ME, Kurtoglu S, Pirgon O, Baykara M. Arterial wall thickening and stiffening in children and adolescents with type 1 diabetes. *Diabetes Res Clin Pract*. 2006;74(1). doi:10.1016/j.diabres.2006.03.004
8. Giannattasio C, Failla M, Piperno A, et al. Early impairment of large artery structure and functionin Type I diabetes mellitus. *Diabetologia*. 1999;42:987-994.
9. Llauradó G, Ceperuelo-Mallafré V, Vilardell C, et al. Arterial stiffness is increased in patients with type 1 diabetes without cardiovascular disease: A potential role of low-grade inflammation. *Diabetes Care*. 2012;35(5):1083-1089. doi:10.2337/dc11-1475
10. Rydén Ahlgren A˚, Sundkvist G, Sandgren T, Länne T, Ahlgren R. *Female Gender Increases Stiffness of Elastic but Not of Muscular Arteries in Type I Diabetic Patients Accepted for Publication*.
11. Kimoto E, Shoji T, Shinohara K, et al. Preferential stiffening of central over peripheral arteries in type 2 diabetes. *Diabetes*. 2003;52(2):448-452. doi:10.2337/diabetes.52.2.448
12. Tanaka H, Palta P, Folsom AR, et al. Habitual Physical Activity and Central Artery Stiffening in Older Adults: The ARIC Study. *J Hypertens*. 2018;36(9):1889-1894. doi:10.1097/HJH.0000000000001782
13. Kozakova M, Palombo C, Mhamdi L, et al. Habitual physical activity and vascular aging in a young to middle-age population at low cardiovascular risk. *Stroke*. Published online 2007. doi:10.1161/STROKEAHA.107.484949
14. Kakiyama T, Matsuda M, Koseki S. Effect of physical activity on the distensibility of the aortic wall in healthy males. *Angiology*. 1998;49(9). doi:10.1177/000331979804901007
15. Peterson MJ, Giuliani C, Morey MC, et al. Physical activity as a preventative factor for frailty: The health, aging, and body composition study. *Journals Gerontol - Ser A Biol Sci Med Sci*. 2009;64(1). doi:10.1093/gerona/gln001

16. Kasapis C, Thompson PD. The effects of physical activity on serum C-reactive protein and inflammatory markers: A systematic review. *J Am Coll Cardiol*. Published online 2005. doi:10.1016/j.jacc.2004.12.077

17. Oh YS. Arterial stiffness and hypertension. *Clin Hypertens*. 2018;24(17).

18. Mercadante AA, Raja A. *Anatomy, Arteries. [Updated 2021 Jan 13]. In: StatPearls [Internet]*. StatPearls Publishing; 2021. Accessed May 25, 2021. https://www.ncbi.nlm.nih.gov/books/NBK547743/?report=reader#_NBK547743_pubdet_

19. DuPont JJ, Kenney RM, Patel AR, Jaffe IZ. Sex differences in mechanisms of arterial stiffness. *Br J Pharmacol*. 2019;176(21):4208-4225. doi:10.1111/bph.14624

20. Majesky MW, Dong XR, Hoglund V, Mahoney WM, Daum G. The adventitia: A dynamic interface containing resident progenitor cells. *Arterioscler Thromb Vasc Biol*. 2011;31(7). doi:10.1161/ATVBAHA.110.221549

21. Nichols WW, Denardo SJ, Wilkinson IB, McEniery CM, Cockcroft J, O'Rourke MF. Effects of arterial stiffness, pulse wave velocity, and wave reflections on the central aortic pressure waveform. *J Clin Hypertens*. 2008;10(4). doi:10.1111/j.1751-7176.2008.04746.x

22. Cecelja M, Chowienczyk P. Role of arterial stiffness in cardiovascular disease. *JRSM Cardiovasc Dis*. Published online 2012. doi:10.1258/cvd.2012.012016

23. Avolio A. Arterial Stiffness. *Pulse*. 2013;1(1):14-28. doi:10.1159/000348620

24. Zieman SJ, Melenovsky V, Kass DA. Mechanisms, pathophysiology, and therapy of arterial stiffness. *Arterioscler Thromb Vasc Biol*. 2005;25(5). doi:10.1161/01.ATV.0000160548.78317.29

25. Xu C, Zarins CK, Pannaraj PS, Bassiouny HS, Glagov S. Hypercholesterolemia superimposed by experimental hypertension induces differential distribution of collagen and elastin. *Arterioscler Thromb Vasc Biol*. 2000;20(12). doi:10.1161/01.ATV.20.12.2566

26. Johnson CP, Baugh R, Wilson CA, Burns J. Age related changes in the tunica media of the vertebral artery: Implications for the assessment of vessels injured by trauma. *J Clin Pathol*. 2001;54(2). doi:10.1136/jcp.54.2.139

27. Harper S. Economic and social implications of aging societies. *Science (80-)*. 2014;346(6209). doi:10.1126/science.1254405

28. Benetos A, Waeber B, Izzo J, et al. Influence of age, risk factors, and cardiovascular and renal disease on arterial stiffness: Clinical applications. *Am J Hypertens*. 2002;15(12). doi:10.1016/S0895-7061(02)03029-7

29. Haluska BA, Jeffries L, Carlier S, Marwick TH. Measurement of arterial distensibility and compliance to assess prognosis. *Atherosclerosis*. 2010;209(2). doi:10.1016/j.atherosclerosis.2009.10.018

30. Mitchell GF, Hwang SJ, Vasan RS, et al. Arterial stiffness and cardiovascular events: The framingham heart study. *Circulation*. Published online 2010. doi:10.1161/CIRCULATIONAHA.109.886655

31. Lakatta EG, Levy D. Arterial and Cardiac Aging: Major Shareholders in Cardiovascular Disease Enterprises Part I: Aging Arteries: A "Set Up" for Vascular Disease The

Demographic Imperative and the Risk of Vascular Diseases in Older Persons. Published online 2003. doi:10.1161/01.CIR.0000048892.83521.58

32. Segers P, Rietzschel ER, Chirinos JA. How to Measure Arterial Stiffness in Humans. *Arterioscler Thromb Vasc Biol*. Published online 2020. doi:10.1161/ATVBAHA.119.313132

33. Bia D, Aguirre I, Zócalo Y, Devera L, Cabrera Fischer E, Armentano R. Regional Differences in Viscosity, Elasticity, and Wall Buffering Function in Systemic Arteries: Pulse Wave Analysis of the Arterial Pressure-Diameter Relationship. *Rev Española Cardiol (English Ed*. 2005;58(2). doi:10.1016/s1885-5857(06)60360-5

34. Wang M, Jiang L, Monticone RE, Lakatta EG. Proinflammation: The key to arterial aging. *Trends Endocrinol Metab*. 2014;25(2). doi:10.1016/j.tem.2013.10.002

35. Deanfield JE, Halcox JP, Rabelink TJ. Endothelial function and dysfunction: Testing and clinical relevance. *Circulation*. Published online 2007:1285-1295. doi:10.1161/CIRCULATIONAHA.106.652859

36. Calles-Escandon J, Cipolla M. Diabetes and endothelial dysfunction: A clinical perspective. *Endocr Rev*. Published online 2001. doi:10.1210/edrv.22.1.0417

37. Quyyumi AA. Endothelial function in health and disease: New insights into the genesis of cardiovascular disease. In: *American Journal of Medicine*. ; 1998. doi:10.1016/s0002-9343(98)00209-5

38. Celermajer DS, Sorensen KE, Spiegelhalter DJ, Georgakopoulos D, Robinson J, Deanfield JE. Aging is associated with endothelial dysfunction in healthy men years before the age-related decline in women. *J Am Coll Cardiol*. 1994;24(2). doi:10.1016/0735-1097(94)90305-0

39. LaRocca TJ, Martens CR, Seals DR. Nutrition and other lifestyle influences on arterial aging. *Ageing Res Rev*. 2017;39. doi:10.1016/j.arr.2016.09.002

40. Ren L, Cai J, Liang J, Li W, Sun H. Impact of cardiovascular risk factors on carotid intima-media thickness and degree of severity: A cross-sectional study. *PLoS One*. Published online 2015. doi:10.1371/journal.pone.0144182

41. Strawbridge RJ, Ward J, Bailey MES, et al. Carotid intima-media thickness novel loci, sex-specific effects, and genetic correlations with obesity and glucometabolic traits in UK Biobank. *Arterioscler Thromb Vasc Biol*. 2020;(February):446-461. doi:10.1161/ATVBAHA.119.313226

42. Tanaka H, Dinenno FA, Monahan KD, DeSouza CA, Seals DR. Carotid artery wall hypertrophy with age is related to local systolic blood pressure in healthy men. *Arterioscler Thromb Vasc Biol*. 2001;21(1):82-87. doi:10.1161/01.ATV.21.1.82

43. Gómez-Marcos MT, Recio-Rodríguez JI, Patino-Alonso MC, et al. Relationship between intima-media thickness of the common carotid artery and arterial stiffness in subjects with and without type 2 diabetes: A case-series report. *Cardiovasc Diabetol*. 2011;10:1-8. doi:10.1186/1475-2840-10-3

44. Coutinho T. Arterial stiffness and its clinical implications in women. *Can J Cardiol*. Published online 2014. doi:10.1016/j.cjca.2014.03.020

45. Zaydun G, Tomiyama H, Hashimoto H, et al. Menopause is an independent factor augmenting the age-related increase in arterial stiffness in the early postmenopausal phase. *Atherosclerosis*. Published online 2006. doi:10.1016/j.atherosclerosis.2005.03.043

46. Hildreth KL, Kohrt WM, Moreau KL. Oxidative stress contributes to large elastic arterial stiffening across the stages of the menopausal transition. *Menopause*. Published online 2014. doi:10.1097/GME.0000000000000116

47. Sztejnsznajd C, da Silva MER, Nussbacher A, et al. Estrogen treatment improves arterial distensibility, fibrinolysis, and metabolic profile in postmenopausal women with type 2 diabetes mellitus. *Metabolism*. 2006;55(7). doi:10.1016/j.metabol.2006.03.003

48. Takato T, Yamada N, Ashida T. Effects of aging and sex on progression of carotid intima-media thickness: A retrospective 6-year follow-up study. *Geriatr Gerontol Int*. Published online 2008. doi:10.1111/j.1447-0594.2008.00467.x

49. Łoboz-Rudnicka M, Jaroch J, Bociąga Z, et al. Impact of cardiovascular risk factors on carotid intima-media thickness: Sex differences. *Clin Interv Aging*. Published online 2016. doi:10.2147/CIA.S103521

50. Laurent S, Cockcroft J, Van Bortel L, et al. Expert consensus document on arterial stiffness: Methodological issues and clinical applications. *Eur Heart J*. 2006;27(21). doi:10.1093/eurheartj/ehl254

51. O'Rourke MF, Staessen JA, Vlachopoulos C, Duprez D, Plante GE. Clinical applications of arterial stiffness; definitions and reference values. *Am J Hypertens*. 2002;15(5). doi:10.1016/S0895-7061(01)02319-6

52. Van Bortel LM, Kool MJ, Struijker Boudier HA. Effects of antihypertensive agents on local arterial distensibility and compliance. In: *Hypertension*. Vol 26. ; 1995. doi:10.1161/01.HYP.26.3.531

53. Godia EC, Madhok R, Pittman J, et al. Carotid artery distensibility: A reliability study. *J Ultrasound Med*. 2007;26(9). doi:10.7863/jum.2007.26.9.1157

54. Selzer RH, Mack WJ, Lee PL, Kwong-Fu H, Hodis HN. Improved common carotid elasticity and intima-media thickness measurements from computer analysis of sequential ultrasound frames. *Atherosclerosis*. 2001;154(1). doi:10.1016/S0021-9150(00)00461-5

55. Yuan C, Wang J, Ying M. Predictive value of carotid distensibility coefficient for cardiovascular diseases and all-cause mortality: A meta-analysis. *PLoS One*. 2016;11(4). doi:10.1371/journal.pone.0152799

56. Kazi AA, Blonde L. *Classification of Diabetes Mellitus. Geneva: World Health Organization; 2019*. Vol 21.; 2019.

57. Leow MKS. Glycated hemoglobin (HbA1c): Clinical applications of a mathematical concept. *Acta Inform Medica*. Published online 2016. doi:10.5455/aim.2016.24.233-238

58. WHO. *Definition and Diagnosis of Diabetes Mellitus and Intermediate Hyperglycemia: Report of a WHO/IDF Consultation*.; 2006.

59. World Health Organization. Report of a World Health Organization Consultation: Use of glycated haemoglobin (HbA1c) in the diagnosis of diabetes mellitus. *Diabetes Res Clin*

Pract. 2011;93(2).

60. Kahn SE, Cooper ME, Del Prato S. Pathophysiology and treatment of type 2 diabetes: Perspectives on the past, present, and future. *Lancet*. 2014;383(9922). doi:10.1016/S0140-6736(13)62154-6

61. Skyler JS, Bakris GL, Bonifacio E, et al. Differentiation of diabetes by pathophysiology, natural history, and prognosis. *Diabetes*. 2017;66(2). doi:10.2337/db16-0806

62. Furutani E. Closed-loop Blood Glucose Control for Type 1 Diabetes. *IEEJ Trans Electron Inf Syst*. 2019;139(4):260-263. doi:10.1541/ieejeiss.139.260

63. Ozougwu, J.C., Okimba, K.C., Belonwu, C.D., Unakalamba CB. The pathogenesis and pathophysiology of type 1 and type 2 diabetes mellitus. *J Physiol Pathophysiol*. Published online 2013. doi:10.5897/jpap2013.0001

64. Shepherd PR, Kahn BB. Glucose transporters and insulin action: Implications for insulin resistance and diabetes mellitus. *N Engl J Med*. Published online 1999. doi:10.1056/NEJM199907223410406

65. Zheng C, Liu Z. Vascular function, insulin action, and exercise: An intricate interplay. *Trends Endocrinol Metab*. Published online 2015. doi:10.1016/j.tem.2015.02.002

66. Alter P, Rupp H, Stoll F, et al. Increased enddiastolic wall stress precedes left ventricular hypertrophy in dilative heart failure - Use of the volume-based wall stress index. *Int J Cardiol*. 2012;157(2). doi:10.1016/j.ijcard.2011.07.092

67. Wang PJ, He F, Liao DH, et al. The impact of age on incremental elastic modulus and incremental compliance of pig hepatic portal vein for liver xenotransplantation. *Xenotransplantation*. 2009;16(1). doi:10.1111/j.1399-3089.2008.00505.x

68. Tynjälä A, Forsblom C, Harjutsalo V, Groop PH, Gordin D. Arterial stiffness predicts mortality in individuals with type 1 diabetes. *Diabetes Care*. 2020;43(9). doi:10.2337/dc20-0078

69. Zeitler P, Hirst K, Pyle L, et al. A Clinical Trial to Maintain Glycemic Control in Youth with Type 2 Diabetes The members of the writing group. *n engl j med*. 2012;24:2247-2256. doi:10.1056/NEJMoa1109333

70. Stumvoll M, Goldstein BJ, Van Haeften TW. Type 2 diabetes: Principles of pathogenesis and therapy. In: *Lancet*. Vol 365. ; 2005. doi:10.1016/S0140-6736(05)61032-X

71. Bogardus C, Lillioja S, Mott DM, Hollenbeck C, Reaven G. Relationship between degree of obesity and in vivo insulin action in man. *Am J Physiol - Endocrinol Metab*. 1985;11(3). doi:10.1152/ajpendo.1985.248.3.e286

72. Mooy JM, Grootenhuis PA, De Vries H, et al. Prevalence and determinants of glucose intolerance in a Dutch Caucasian population: The Hoorn study. *Diabetes Care*. 1995;18(9). doi:10.2337/diacare.18.9.1270

73. World Health Organization W. *Classification of Diabetes Mellitus. Geneva: World Health Organization; 2019. Licence: CC BY-NC-SA 3.0 IGO.*; 2019.

74. Cameron JD, Bulpitt CJ, Pinto ES, Rajkumar C. The aging of elastic and muscular arteries: A comparison of diabetic and nondiabetic subjects. *Diabetes Care*. 2003;26(7).

doi:10.2337/diacare.26.7.2133

75. Charvat J, Chlumsky J, Zakovicova E, Kvapil M. *Common Carotid Artery Intima-Media Thickness Is Not Increased but Distensibility Is Reduced in Normotensive Patients with Type 2 Diabetes Compared with Control Subjects*. Vol 38.; 2010.

76. Huang PL. A comprehensive definition for metabolic syndrome. *DMM Dis Model Mech*. 2009;2(5-6). doi:10.1242/dmm.001180

77. Park S, Lakatta EG. Role of inflammation in the pathogenesis of arterial stiffness. *Yonsei Med J*. 2012;53(2):258-261. doi:10.3349/ymj.2012.53.2.258

78. Nakhai-Pour HR, Grobbee DE, Bots ML, Muller M, Van der Schouw YT. C-reactive protein and aortic stiffness and wave reflection in middle-aged and elderly men from the community. *J Hum Hypertens*. 2007;21(12):949-955. doi:10.1038/sj.jhh.1002255

79. King DE, Mainous AG, Buchanan TA, Pearson WS. C-reactive protein and glycemic control in adults with diabetes. *Diabetes Care*. 2003;26(5):1535-1539. doi:10.2337/diacare.26.5.1535

80. Benigni A, Cassis P, Remuzzi G. Angiotensin II revisited: New roles in inflammation, immunology and aging. *EMBO Mol Med*. Published online 2010. doi:10.1002/emmm.201000080

81. Little RR, Sacks DB. HbA1c: How do we measure it and what does it mean? *Curr Opin Endocrinol Diabetes Obes*. 2009;16(2). doi:10.1097/MED.0b013e328327728d

82. Selvin E, Marinopoulos S, Berkenblit G, et al. Meta-analysis: Glycosylated hemoglobin and cardiovascular disease in diabetes mellitus. *Ann Intern Med*. 2004;141(6). doi:10.7326/0003-4819-141-6-200409210-00007

83. Zhang Y, Hu G, Yuan Z, Chen L. Glycosylated hemoglobin in relationship to cardiovascular outcomes and death in patients with type 2 diabetes: A systematic review and meta-analysis. *PLoS One*. 2012;7(8). doi:10.1371/journal.pone.0042551

84. Mannucci E, Dicembrini I, Lauria A, Pozzilli P. Is glucose control important for prevention of cardiovascular disease in diabetes? *Diabetes Care*. 2013;36(SUPPL.2). doi:10.2337/dcS13-2018

85. Moreno B, De Faria AP, Ritter AMV, et al. Glycated hemoglobin correlates with arterial stiffness and endothelial dysfunction in patients with resistant hypertension and uncontrolled diabetes mellitus. *J Clin Hypertens*. 2018;20(5). doi:10.1111/jch.13293

86. Prasad K. C-Reactive Protein and Cardiovascular Diseases. doi:10.1007/s00547-003-1018-y

87. Shrivastava AK, Singh HV, Raizada A, Singh SK. C-reactive protein, inflammation and coronary heart disease. *Egypt Hear J*. 2015;67(2). doi:10.1016/j.ehj.2014.11.005

88. McEniery CM, Wallace S, Mackenzie IS, Cockcroft JR, Wilkinson IB. C-Reactive Protein Is Associated With Arterial Stiffness in Apparently Healthy Individuals. Published online 2004. doi:10.1161/01.ATV.zhq0504.0173

89. Mattace-Raso FUS, Van Der Cammen TJM, Van Der Meer IM, et al. C-reactive protein and arterial stiffness in older adults: The Rotterdam Study. *Atherosclerosis*. 2004;176(1).

doi:10.1016/j.atherosclerosis.2004.04.014

90. Anderson TJ, Grégoire J, Pearson GJ, et al. 2016 Canadian Cardiovascular Society Guidelines for the Management of Dyslipidemia for the Prevention of Cardiovascular Disease in the Adult. *Can J Cardiol*. 2016;32(11). doi:10.1016/j.cjca.2016.07.510

91. Talayero BG, Sacks FM. The role of triglycerides in atherosclerosis. *Curr Cardiol Rep*. 2011;13(6). doi:10.1007/s11886-011-0220-3

92. Wang X, Ye P, Cao R, et al. Triglycerides are a predictive factor for arterial stiffness: a community-based 4.8-year prospective study. Published online 2016. doi:10.1186/s12944-016-0266-8

93. Alexopoulos AS, Qamar A, Hutchins K, Crowley MJ, Batch BC, Guyton JR. Triglycerides: Emerging Targets in Diabetes Care? Review of Moderate Hypertriglyceridemia in Diabetes. *Curr Diab Rep*. 2019;19(4). doi:10.1007/s11892-019-1136-3

94. Lemieux I, Lamarche B, Couillard C, et al. Total cholesterol/HDL cholesterol ratio vs LDL cholesterol/HDL cholesterol ratio as indices of ischemic heart disease risk in men. *Arch Intern Med*. 2001;161(22). doi:10.1001/archinte.161.22.2685

95. Wang F, Ye P, Luo L, et al. Association of serum lipids with arterial stiffness in a population-based study in Beijing. *Eur J Clin Invest*. 2011;41(9). doi:10.1111/j.1365-2362.2011.02481.x

96. Ference BA, Ginsberg HN, Graham I, et al. Low-density lipoproteins cause atherosclerotic cardiovascular disease. 1. Evidence from genetic, epidemiologic, and clinical studies. A consensus statement fromthe European Atherosclerosis Society Consensus Panel. *Eur Heart J*. 2017;38(32). doi:10.1093/eurheartj/ehx144

97. Barter P. The role of HDL-cholesterol in preventing atherosclerotic disease. In: *European Heart Journal, Supplement*. Vol 7. ; 2005. doi:10.1093/eurheartj/sui036

98. Jukema JW, Liem A-H, Dunselman PHJM, et al. LDL-C/HDL-C ratio in subjects with cardiovascular disease and a low HDL-C: results of the RADAR (Rosuvastatin and Atorvastatin in different Dosages And Reverse cholesterol transport) study LDL-C/HDL-C ratio in subjects with cardiovascular disease and a low HDL-C: results of the RADAR (Rosuvastatin and Atorvastatin in different Dosages Atorvastatin in different Dosages And Reverse cholesterol And Reverse cholesterol transport) study. 2005;21(11):1865-1874. doi:10.1185/030079905X74952

99. Prince CT, Secrest AM, Mackey RH, Arena VC, Kingsley LA, Orchard TJ. Cardiovascular autonomic neuropathy, HDL cholesterol, and smoking correlate with arterial stiffness markers determined 18 years later in type 1 diabetes. *Diabetes Care*. 2010;33(3). doi:10.2337/dc09-1936

100. Cao Y, Yan L, Guo N, et al. Non-high-density lipoprotein cholesterol and risk of cardiovascular disease in the general population and patients with type 2 diabetes: A systematic review and meta-analysis. *Diabetes Res Clin Pract*. 2019;147. doi:10.1016/j.diabres.2018.11.002

101. Colhoun HM, Betteridge DJ, Durrington PN, et al. Primary prevention of cardiovascular

disease with atorvastatin in type 2 diabetes in the Collaborative Atorvastatin Diabetes Study (CARDS): Multicentre randomised placebo-controlled trial. *Lancet*. 2004;364(9435). doi:10.1016/S0140-6736(04)16895-5

102. Flack JM, Adekola B. Blood pressure and the new ACC/AHA hypertension guidelines. *Trends Cardiovasc Med*. 2020;30(3). doi:10.1016/j.tcm.2019.05.003

103. Safar ME. Systolic blood pressure, pulse pressure and arterial stiffness as cardiovascular risk factors. *Curr Opin Nephrol Hypertens*. 2001;10(2). doi:10.1097/00041552-200103000-00015

104. DHHS. National Diabetes Statistics Report, 2020. *Natl Diabetes Stat Rep*. Published online 2020.

105. Forouzanfar M, Dajani HR, Groza VZ, Bolic M, Rajan S, Batkin I. Oscillometric blood pressure estimation: Past, present, and future. *IEEE Rev Biomed Eng*. 2015;8. doi:10.1109/RBME.2015.2434215

106. Rutan GH, Kuller LH, Neaton JD, Wentworth DN, McDonald RH, McFate Smith W. Mortality associated with diastolic hypertension and isolated systolic hypertension among men screened for the Multiple Risk Factor Intervention Trial. *Circulation*. 1988;77(3). doi:10.1161/01.CIR.77.3.504

107. Strandberg TE, Pitkala K. What is the most important component of blood pressure: Systolic, diastolic or pulse pressure? *Curr Opin Nephrol Hypertens*. 2003;12(3). doi:10.1097/00041552-200305000-00011

108. Pinto E. Blood pressure and ageing. *Postgrad Med J*. 2007;83(976). doi:10.1136/pgmj.2006.048371

109. Webb AJS. Progression of Arterial Stiffness is Associated With Midlife Diastolic Blood Pressure and Transition to Late-Life Hypertensive Phenotypes. *J Am Heart Assoc*. 2020;9(1). doi:10.1161/JAHA.119.014547

110. Grossman E. Does increased oxidative stress cause hypertension? *Diabetes Care*. 2008;31 Suppl 2. doi:10.2337/dc08-s246

111. Dharmashankar K, Widlansky ME. Vascular endothelial function and hypertension: Insights and directions. *Curr Hypertens Rep*. 2010;12(6). doi:10.1007/s11906-010-0150-2

112. Schulz E, Gori T, Münzel T. Oxidative stress and endothelial dysfunction in hypertension. *Hypertens Res*. 2011;34(6). doi:10.1038/hr.2011.39

113. Nuttall FQ. Body mass index: Obesity, BMI, and health: A critical review. *Nutr Today*. 2015;50(3). doi:10.1097/NT.0000000000000092

114. Khan SS, Ning H, Wilkins JT, et al. Association of Body Mass Index With Lifetime Risk of Cardiovascular Disease and Compression of Morbidity Supplemental content. *JAMA Cardiol*. 2018;3(4):280-287. doi:10.1001/jamacardio.2018.0022

115. Strasser B, Arvandi M, Pasha EP, Haley AP, Stanforth P, Tanaka H. Abdominal obesity is associated with arterial stiffness in middle-aged adults. *Nutr Metab Cardiovasc Dis*. 2015;25(5). doi:10.1016/j.numecd.2015.01.002

116. Carbone S, Canada JM, Billingsley HE, Siddiqui MS, Elagizi A, Lavie CJ. Obesity

paradox in cardiovascular disease: Where do we stand? *Vasc Health Risk Manag*. 2019;15. doi:10.2147/VHRM.S168946

117. Tanaka H, DeSouza CA, Seals DR. Absence of age-related increase in central arterial stiffness in physically active women. *Arterioscler Thromb Vasc Biol*. Published online 1998. doi:10.1161/01.ATV.18.1.127

118. Schmitz KH, Arnett DK, Bank A, et al. Arterial distensibility and physical activity in the ARIC study. *Med Sci Sports Exerc*. 2001;33(12). doi:10.1097/00005768-200112000-00014

119. Minder CM, Shaya GE, Michos ED, et al. Relation between self-reported physical activity level, fitness, and cardiometabolic risk. *Am J Cardiol*. 2014;113(4). doi:10.1016/j.amjcard.2013.11.010

120. Moreau KL, Silver AE, Dinenno FA, Seals DR. Habitual aerobic exercise is associated with smaller femoral artery intima media thickness with age in healthy men and women. *Eur J Prev Cardiol*. Published online 2006. doi:10.1097/01.hjr.0000230103.55653.42

121. Gando Y, Yamamoto K, Kawano H, et al. Attenuated age-related carotid arterial remodeling in adults with a high level of cardiorespiratory fitness. *J Atheroscler Thromb*. Published online 2011. doi:10.5551/jat.6924

122. Kozakova M, Palombo C, Mhamdi L, et al. Habitual physical activity and vascular aging in a young to middle-age population at low cardiovascular risk. *Stroke*. Published online 2007. doi:10.1161/STROKEAHA.107.484949

123. Baecke JAH, Burema J, Frijters JER. A short questionnaire for the measurement of habitual physical activity in epidemiological studies. *Am J Clin Nutr*. 1982;36(5). doi:10.1093/ajcn/36.5.936

124. Cotie LM, Josse AR, Philips SM, MacDonald MJ. 16-Weeks of Combined Aerobic and Resistance Training and Hypo-Caloric Diet on Measures of Arterial Stiffness in Overweight Pre-Menopausal Women. *J Metab Syndr*. 2014;03(01). doi:10.4172/2167-0943.1000137

125. Cotie LM, Currie KD, McGill GM, et al. Associations between measures of vascular structure and function and systemic circulating blood markers in humans. *Physiol Rep*. 2016;4(18). doi:10.14814/phy2.12982

126. Königstein K, Infanger D, Klenk C, Carrard J, Hinrichs T, Schmidt-Trucksäss A. Physical activity is favorably associated with arterial stiffness in patients with obesity and elevated metabolic risk. *Int J Clin Pract*. 2020;74(9). doi:10.1111/ijcp.13563

127. Stamatelopoulos K, Tsoltos N, Armeni E, et al. Physical activity is associated with lower arterial stiffness in normal-weight postmenopausal women. *J Clin Hypertens*. 2020;22(9). doi:10.1111/jch.13954

128. Raina PS, Wolfson C, Kirkland SA, et al. The canadian longitudinal study on aging (CLSA). *Can J Aging*. 2009;28(3):221-229. doi:10.1017/S0714980809990055

129. Raina P, Wolfson C, Kirkland S, et al. Cohort Profile: The Canadian Longitudinal Study on Aging (CLSA). *Int J Epidemiol*. 2019;48(6):1752-1753J. doi:10.1093/ije/dyz173

130. Raina P, Wolfson C, Kirkland S, Griffith L, Griffi L. The Canadian Longitudinal Study on Aging (CLSA) Report on Health and Aging in Canada - Findings from Baseline Data Collection. Published online 2018. https://www.clsa-elcv.ca/doc/2639

131. Maintaining Contact Questionnaire (Tracking and Comprehensive): CLSA Scientific Working Groups. Published online 2015. https://clsa-elcv.ca/doc/540

132. Washburn RA, Smith KW, Jette AM, Janney CA. The physical activity scale for the elderly (PASE): Development and evaluation. *J Clin Epidemiol*. 1993;46(2). doi:10.1016/0895-4356(93)90053-4

133. Choi HL, Au JS, MacDonald MJ. Carotid extra-media thickness increases with age, but is not related to arterial stiffness in adults. *Artery Res*. 2018;21. doi:10.1016/j.artres.2017.12.003

134. Kamruzzaman MD, M Rahmatullah Imon AH. High leverage point: another source of multicollinearity. *Pakistan J Stat.* 2002;(January 2002).

135. Cook RD, Weisberg S. Residuals and Influence in Regression. In: ; 1982.

136. Iacobucci D, Schneider MJ, Popovich DL, Bakamitsos GA. Mean centering helps alleviate "micro" but not "macro" multicollinearity. *Behav Res Methods*. 2016;48(4). doi:10.3758/s13428-015-0624-x

137. Iacobucci D, Schneider MJ, Popovich DL, Bakamitsos GA. Mean centering, multicollinearity, and moderators in multiple regression: The reconciliation redux. *Behav Res Methods*. 2017;49(1). doi:10.3758/s13428-016-0827-9

138. Tsuchikura S, Shoji T, Kimoto E, et al. Central versus peripheral arterial stiffness in association with coronary, cerebral and peripheral arterial disease. *Atherosclerosis*. 2010;211(2). doi:10.1016/j.atherosclerosis.2010.03.037

139. Logan SL, Gottlieb BH, Maitl SB, Meegan D, Spriet LL. The physical activity scale for the elderly (PASE) questionnaire; Does it predict physical health? *Int J Environ Res Public Health*. Published online 2013. doi:10.3390/ijerph10093967

140. Smulyan H, Lieber A, Safar ME. Hypertension, Diabetes Type II, and their association: Role of arterial stiffness. *Am J Hypertens*. 2016;29(1). doi:10.1093/ajh/hpv107

141. Fox CS, Sullivan L, D'Agostino RB, Wilson PWF. The Significant Effect of Diabetes Duration on Coronary Heart Disease Mortality: The Framingham Heart Study. *Diabetes Care*. 2004;27(3). doi:10.2337/diacare.27.3.704

142. Spijkerman AMW, Dekker JM, Nijpels G, et al. Impact of diabetes duration and cardiovascular risk factors on mortality in type 2 diabetes: The Hoorn Study. *Eur J Clin Invest*. 2002;32(12). doi:10.1046/j.1365-2362.2002.01090.x

143. Vaidya D, Heckbert SR, Wasserman BA, Ouyang P. Sex-specific association of age with carotid artery distensibility: Multi-ethnic study of atherosclerosis. *J Women's Heal*. 2012;21(5). doi:10.1089/jwh.2011.3220

144. De Angelis L, Millasseau SC, Smith A, et al. Sex differences in age-related stiffening of the aorta in subjects with type 2 diabetes. *Hypertension*. 2004;44(1). doi:10.1161/01.HYP.0000130482.81883.fd

145. Mitchell GF. Arterial stiffness and hypertension: Chicken or egg? *Hypertension*. 2014;64(2). doi:10.1161/HYPERTENSIONAHA.114.03449

146. McGorrian C, Yusuf S, Islam S, et al. Estimating modifiable coronary heart disease risk in multiple regions of the world: The INTERHEART Modifiable Risk Score. *Eur Heart J*. 2011;32(5). doi:10.1093/eurheartj/ehq448

147. Yusuf S, Rangarajan S, Teo K, et al. Cardiovascular Risk and Events in 17 Low-, Middle-, and High-Income Countries. *N Engl J Med*. 2014;371(9). doi:10.1056/nejmoa1311890

148. Privšek E, Hellgren M, Råstam L, Lindblad U, Daka B. Epidemiological and clinical implications of blood pressure measured in seated versus supine position. *Med (United States)*. 2018;97(31). doi:10.1097/MD.0000000000011603

CHAPTER 2: MANUSCRIPT

2.1 Introduction

The global population is aging, and it is expected that the number of adults over the age of 65 will double by the year 2050 [27]. Aging has been identified as an independent risk factor for developing cardiovascular disease (CVD), which presents a major health concern[3,4]. CVDs include diseases of the heart and vasculature, such as coronary heart disease, heart failure, peripheral arterial disease, and cerebrovascular disease [1,2]. CVDs are considered one the leading causes of death, accounting for 31% of deaths worldwide [1]. The American Heart Association reported that CVD incidence in the US is ~40% between 40–59 years of age, ~75% between 60–79 years of age, and ~86% in those over age 80 [3].

It is well established that aging is associated with increases in arterial stiffness, which is the progressive impairment of an artery to constrict and dilate in response to changes in blood pressure [22,28]. The structural integrity and compliance of the arterial wall are partially dependent upon the relative contribution of the scaffolding proteins elastin and collagen [24]. The balance of these structural proteins is maintained through repeated cycles of production and degradation [24]. Over time, arterial walls thicken and lose elasticity, resulting in an impaired buffering capacity of the central conduit arteries, whose elastic properties ensure anterograde blood flow during diastole [33]. With losses in elasticity, to ensure adequate tissue level blood flow despite impaired elastic energetic contribution during diastole increased flow during systole is required, thereby necessitating higher systolic pressures[32]. These elevated systolic pressures result in greater cardiac work and mechanical strain on the downstream blood vessels, organs, and tissues. Sex

differences exist in the age-related increase in arterial stiffness, such that arterial stiffness and the risk of cardiovascular (CV) events both increase drastically in females following menopause compared to males of the same age [44]. Aging has been identified as an independent predictor of CVD, and the presence of additional risk factors associated with aging, including hypertension, obesity, inflammation, and diabetes, can exacerbate that risk, [4].

Diabetes mellitus (DM) is a metabolic disorder characterized by chronic hyperglycemia resulting from impaired secretion and/or action of insulin [5]. While it is considered an independent risk factor for developing CVD, DM can also be a precursor to hypertension, obesity, and chronic inflammation [5]. DM is typically either Type 1 or Type 2, however less common forms such as gestational and monogenic diabetes also exist [59–61]. Type 1 diabetes mellitus (T1DM), also known as insulin-dependent diabetes mellitus, is a chronic autoimmune disorder characterized by the selective destruction of insulin-producing pancreatic beta cells [62,63]. Type 2 diabetes mellitus (T2DM) is characterized by insulin resistance and other health complications such as abdominal obesity, hypertension, dyslipidemia and hyperglycemia [5,60]. In contrast with T1DM, most people with T2DM are either overweight or obese, which either causes or exacerbates, their resistance to insulin [5,70,71]. Common across diabetes type is accelerated vascular stiffening, such that individuals with DM typically display high arterial stiffness relative to healthy, non-diabetic counterparts [7–9,11,74].

Individuals who are more physically active and/or those with a higher degree of physical fitness may have attenuated age-induced arterial stiffening [12,13,117–121]. Objectively measured habitual physical activity (PA) is negatively associated with arterial stiffness in healthy older adults [13,117]. The likely mechanism by which habitual PA attenuates age-related arterial stiffening is *via* downregulation of pro-inflammatory cytokine production in adipose and muscle tissue and

upregulation of atheroprotective cytokine signalling, resulting in a cardio-protective effect [16]. However, findings from individuals with an elevated cardiometabolic risk profile are not as conclusive [126,127]. Königstein *et al.* (2020) recently demonstrated that reduced brachial-ankle pulse wave velocity (baPWV), an indicator of decreased arterial stiffness, was strongly associated with objectively measured increases in PA in 55 middle-aged adults (43.0 ± 13.8 years; 66% women) with obesity. Of these individuals, 20 had T2DM, and only moderate-vigorous intensity PA (MVPA), but not light-intensity PA (LPA) or cardiorespiratory fitness, was associated with lower baPWV[126]. In contrast, Stamatelopoulos *et al.* (2020) recently demonstrated that subjectively measured PA (using a modified version of the International Physical Activity Questionnaire) was inversely associated with carotid-femoral pulse wave velocity (cfPWV), the current gold standard for arterial stiffness measurement, in normal weight, but not obese, post-menopausal women (mean age 57.7 ± 7.6 years) supporting the concept that increased PA was associated with reduced arterial stiffness. This discrepancy between previous studies highlights the need to examine the influence of subjectively measured PA on central arterial stiffness in an older group of adults with an elevated cardiometabolic risk and the also need to include both biological sexes in these examinations.

What is currently unknown is whether self-reported PA is associated with accelerated arterial stiffness in older Canadians with T2DM and whether differences between sexes exist in this population. The specific objectives of this study were to: evaluate the influence of self-reported PA on arterial stiffness in older adults with T2DM and other DM2O while controlling for known CVD risk factors and markers, and to assess whether this relationship is modulated by age and sex. It was hypothesized that, similar to healthy older adults, more physically active older individuals with T2DM would have attenuated arterial stiffness compared to their more

sedentary counterparts. Furthermore, we hypothesized that sex differences would exist in this relationship, such that the association with PA will be attenuated in females with DM2O.

2.2 Methods

2.2.1 Study Design: CLSA Data Collection Methods

The Canadian Longitudinal Study on Aging (CLSA) consists of a national stratified random sample of 51,338 Canadian men and women between the ages of 45-85 at baseline [128,129]. This sample is further delineated into two cohorts: a "Tracking" cohort and a "Comprehensive" cohort [128,129]. The Tracking cohort comprises 21,241 individuals randomly selected from the 10 Canadian provinces who provided questionnaire data via computer-assisted telephone interview (CATI) only[128,129]. The Comprehensive cohort comprises 30,097 participants randomly selected within 25-50 kilometres of data collection sites (Vancouver, Victoria, Calgary, Winnipeg, Hamilton, Ottawa, Montreal, Sherbrooke, Halifax, and St. Johns) [128–130]. Data from the Comprehensive cohort was collected via detailed computer-assisted personal interviewing (CAPI), physical assessments and biological (blood and urine) samples [128–130]. Baseline data for the Tracking cohort was collected between 2011 and 2014, and baseline data for the Comprehensive cohort was collected between 2012 and 2015 [130]. Data will be collected every three years for 20 years, or until participant death or withdrawal from the study [128]. The Maintaining Contact questionnaire was administered 18 months after baseline via CATI software to both the Tracking and Comprehensive cohort [131]. Physical activity data was only collected *via* the Maintaining Contact questionnaire, but the remainder of the variables in the present study are derived from baseline questionnaires, physical assessments, and biological samples from the Comprehensive cohort. Ethical clearance for this project was obtained from the McMaster

Research Ethics Board, and permission to access CLSA data was obtained *via* the Co-Applicant Agreement Form for data access.

2.2.2 Participants

Of the 30,097 participants in the CLSA Comprehensive cohort, 5,310 participants self-identified as having diabetes. The specific cohort of interest for this study includes individuals who self-identified as having diabetes. These individuals answered, 'yes' to the question "has a doctor ever told you that you have diabetes?" from the CLSA baseline questionnaire. Any missing values or respondents who answered "I do not know" were excluded. Of these individuals, 4,666 had complete biological samples taken and had ultrasound image files available for analysis. Of these 4,666 individuals, a total of 2,201 individuals had ultrasound image files that met the quality assessment criteria to be analyzed for arterial distensibility and were included in the final cohort.

Physical Activity Scale for the Elderly (PASE)

Physical activity was assessed using a modified version Physical Activity Scale for the Elderly (PASE), a brief and easily scored 12-item survey used to assess usual physical activity in adults 65 years and older [132]. This scale is specifically designed for elderly individuals and this includes questions and activities tailored for this population. These activities include sitting, walking, light, moderate or strenuous sports/recreational activities, muscular strength/endurance activities, housework, home repairs, yard work and gardening, caring for others, and working/volunteering [131]. The frequency, duration, and intensity level of these activities over the seven days before reporting are used to generate a score between 0 to 793. Higher scores indicate greater levels of physical activity. It has been demonstrated that this is a valid and reliable metric for assessing physical activity in elderly populations [132].

Blood Pressure

As per the CLSA heart rate and blood pressure data collection protocol version 2.2, blood pressure was recorded on the left side of the body using an automatic blood pressure machine (BpTRU Vital Signs Monitor, Coquitlam, Canada). A total of six measurements were taken, and the systolic and diastolic blood pressure used in the present study was the average of those measurements, excluding the first reading. Pulse pressure was calculated as the difference between the average systolic and diastolic blood pressures.

2.2.2 Data Analysis

Ultrasound Image Analysis

As per the CLSA data collection protocol for cIMT version 4.1, participants were asked to lie in a supine position for 5-10 minutes prior to beginning the test. During this time, participants were instrumented with three electrodes to assess the electrical activity of the heart via electrocardiogram (ECG) (GE MAC 1600 ECG Analysis System). Following the 5–10-minute rest period, longitudinal brightness mode video clips of 3 consecutive heart cycles (cineloops) were captured from images obtained at a depth of 4 cm from the skin surface at the site of the common carotid artery bifurcation using an 11Mhz probe connected to a commercial ultrasound unit (VIVIDi; GE Medical Systems, Horten, Norway). The cineloops were analyzed to calculate the average posterior wall thickness (IMT) of both the right and left carotid arteries. The cineloop considered the best quality was labelled a “parent” file, meaning the cIMT value associated with that participant is derived from that cineloop. Any other cineloops are considered “childless.” cIMT was also calculated during the arterial stiffness analysis protocol, expressed as an average of the far wall IMT over the 3 heart cycles.

The same ultrasound images (cineloops) were stored on external hard drives in Digital Image and Communications in Medicine (DICOM) format to determine arterial stiffness calculated as arterial distensibility. Distensibility was calculated using the equation below:

$$(1)\ \text{Distensibility} = \frac{\Delta\text{Cross- Sectional Area}}{\text{Pulse Pressure} \times \text{Minimum Cross- Sectional Area}}$$

Wherein CSA is the difference between the maximum CSA and minimum CSA, and pulse pressure is the difference between resting systolic and diastolic blood pressure. Distensibility represents the relative change in arterial diameter for a given change in pulse pressure [51]. Image analysis was conducted using a semi-automated edge-detection software [AMS (Artery Measurement System) Image and Data Analysis; Gothenburg, Sweden] used to detect the walls of the carotid artery lumen and intima-media based on contrasting brightness intensities. A total of 6 frames were captured at an estimated frame rate of 27 frames per second (fps). A single cardiac cycle consisted of two frames, which corresponded to the minimum diastolic and peak systolic diameters, respectively, as calculation of arterial distensibility requires the minimum and maximum arterial diameters [51]. A region of interest was selected based on clarity of artery and far-arterial wall IMT, and the quality of all 6 frames needed to be considered. Prior to image analysis, a quality control protocol was performed to ensure the image was analyzable for distensibility. Images did not pass quality control if the artery was not visible, the far wall cIMT was not visible, or nothing was identifiable for 4 of 6 frames. Images were also not analyzed if the ECG data was incorrectly recorded or not present as it is required to determine systole and diastole. If the cineloop passed quality control, each frame was scanned for the region of interest placement and any necessary manual edge-detection adjustments were made [133]. Minimum, maximum, and mean lumen diameters, as well as mean IMT of the opposite arterial wall, were calculated for each frame.

Diabetes Classification

Individuals were stratified based on diabetes type based on self-reported data from the data collection site interview questionnaire, in which participants were asked the type of diabetes with which they were previously diagnosed. The possible responses were: "Type 1, Type 2, neither, don't know/no answer, and refused." For this study, diabetes type was considered either "Type 1" or "DM2O" which encompasses Type 2, gestational diabetes, and other variants that were not discernable within this data set, including individuals who indicated they had been diagnosed with diabetes, but when asked about the type, indicated "neither," or "don't know/no answer." Type 1 diabetes is an autoimmune condition characterized by the destruction of insulin-producing pancreatic beta cells, resulting in insulin deficiency [5]. Individuals with Type 1 diabetes require exogenous insulin to maintain healthy blood glucose levels [62]. Four individuals who indicated that they had been diagnosed with type 1 diabetes were re-distributed to the "DM2O" category based on current classification for type 1 diabetes. All four individuals indicated that they were not currently taking any form of drug for their diabetes and did not indicate that they were taking any form of insulin. Two were considered class 2 obese based on BMI, and three had an age of diagnosis above 40 years of age [5]. To allow for greater specificity in the interpretation and generalizability of results, and to align with the purpose of this research project, individuals with Type 1 diabetes were also removed from the final model.

2.2.3 Statistical analysis

Statistical analysis was performed using IBM SPSS Statistics for MacOS (version 28.0; Chicago, IL). All variables were assessed for normality before data analysis. Normality of data was assessed via visual assessment of the distribution, assessing the range of skewness and kurtosis, and the Shapiro-Wilks test for normality. The main dependent variable of interest,

arterial distensibility, was non-normal. For variables with missing data, a paired samples t-test was conducted to determine whether any difference was present between cases with a missing variable and cases without a missing value for the main outcome variable. To assess the impact of missing data, variables with missing data were dummy coded to allow for comparison between those with data and those without data on the main outcome variable of interest to ensure data was missing at random.

A multiple linear regression within the general linear model analysis was performed to assess the influence of PASE score on arterial distensibility adjusting only for age and sex. Covariates used in the adjusted model included: smoking status, self-declared heart disease, systolic blood pressure (SBP), diastolic blood pressure (DBP), carotid intima media thickness (IMT), high-sensitivity C-reactive protein (hsCRP), glycated hemoglobin (HbA_{1c}), triglycerides, high-density lipoprotein cholesterol (HDL-C), and low-density lipoprotein cholesterol (LDL-C). Data were assessed for multicollinearity via the correlation matrix. Total cholesterol was not included due to multicollinearity with LDL-C. The initial model generated a studentized residual score for distensibility. Outliers were identified as a studentized residual score greater, or less than, 3, or -3 respectively, reflective of a studentized residual score greater than three standard deviations away from the mean for distensibility. Linearity of the data was assessed *via* visual assessment of a scatter plot of the studentized residual and unstandardized predicted value. All leverage values (a measure of extreme predictor *x* values, used to determine outliers and multicollinearity) were found to be below 0.2 [134]. Cook's distance was assessed, but no values were above 1[135]. Continuous variables were mean-centred before building the regression model to clarify the interpretation of regression coefficients without altering $R^{2;}$ however, this may reduce multicollinearity in regression analyses that include interaction terms [136,137].

2.3 Results

2.3.1 Participants Characteristics

General participant characteristics are presented in Table 1. Characteristics are presented stratified by sex in Table 2, and Table 3 displays participants' characteristics stratified by diabetes type. Comparison of means in Table 2 and Table 3 are presented *via* independent sample t-tests or Mann-Whitney U test, depending on the normality of the variable. Table 4 provides a breakdown of the sample by diabetes type and sex relative to the sample included in the analysis of the present study (n=2,201) and the entire comprehensive cohort of the CLSA (N = 30,097). There were 75 individuals with T1DM, which accounted for only 3.4% of the sample (n = 2,201). Table 5 presents the available self-reported hypertension characteristics, stratified by diabetes type and sex. 56% of the sample self-reported a previous diagnosis of hypertension, and 47% of the total sample indicated that they were taking anti-hypertensive medication at the time of data collection. Table 6 presents the available self-reported heart disease data stratified by sex and diabetes type. 17% of the sample self-reported a previous diagnosis of heart disease. Table 7 presents the correlation matrix for the independent correlations between the dependent variable (arterial distensibility) and each independent variable in the entire cohort (n= 2,201). Correlational analysis via Spearman's Rho revealed a weak positive correlation between PASE and arterial distensibility (0.151, *P*=<.001). Table 8 presents the correlation matrix for the independent correlations between the dependent variable (arterial distensibility) and each independent variable in the DM2O cohort (n= 2,126).

2.3.2 Influence of PASE Score on Arterial Distensibility

The initial unadjusted model with possible interactions included 2,003 individuals. R^2 for the overall model was 0.086, while the adjusted R^2 was 0.084 (Table 9). No significant

interactions were noted between age and sex, age and PASE, or sex and PASE. The second unadjusted model with outliers and non-significant interactions removed included 1,979 individuals. R^2 for the overall model was 0.107, while the adjusted R^2 was 0.106 (Table 10). The final sample included in a general linear model was 1,727. R^2 for the overall model was 0.248%, while the adjusted R^2 was 0.242% (Table 11).

The main finding of the present study is that there was no main effect of PASE score on arterial distensibility before (P = 0.143) and after (P = 0.998) adjusting for known CV risk factors and markers. There was a main effect of age on arterial distensibility in both models (P=<0.001). The addition of the covariates in the final adjusted model allowed for the small but significant detection of a main effect of age (P=0.040). Main effects of the other independent variables in the model are presented in Table 9. Notably, no main effect was found for hsCRP on arterial distensibility (P=0.595).

Table 1: Sample Characteristics

	Total (n = 2201)	
	Mean (SD)	***Median (IQR)***
Age (years)	65 (10)	65 (58-72)
Age of Diagnosis (years) [a, b]	56 (14)	57 (49-65)
Distensibility ($mmHg^{-1}$) [b]	0.0022 (.0010)	0.0020 (.0014-.0027)
cIMT (mm)	0.77 (0.17)	0.74 (0.64-0.86)
BMI [a]	29.5 (5.2)	28.9 (25.7-32.4)
hs-CRP (mg/L) [b]	2.8 (5.3)	1.3 (.7-2.9)
HbA1c (%) [b]	6.4 (1.2)	6.1 (5.6-6.8)
Triglycerides (mM) [b]	1.99 (1.11)	1.7 (1.22-2.46)
HDL (mM) [b]	1.35 (.44)	1.28 (1.03-1.59)
LDL (mM) [a]	2.41 (.99)	2.25 (1.66-3.09)
Cholesterol (mM)	4.65 (1.18)	4.50 (3.75-5.42)
TC/HDL Ratio [b]	3.70 (1.33)	3.44 (2.76-4.30)
SBP (mmHg)	123.4 (16.2)	121.6 (112.6-132.7)
DBP (mmHg)	73.4 (10.0)	73.4 (66.3-79.6)
Maximal Grip Strength (kg) [a]	34.93 (11.18)	33.86 (25.93-42.82)
PASE Score [a, b]	131.4 (69.3)	119.6 (82.1-171.0)
Get Up and Go Time (seconds) [a, b]	10.04 (2.39)	9.65 (8.53-11.03)
4 Metre Walk Time (seconds) [a, b]	4.42 (1.04)	4.22 (3.72-4.91)
Total Chair Rise Time (seconds) [a, b]	13.87 (3.73)	13.40 (11.37-15.82)

[a] Variable was missing for less than 9% of participants; [b] Data was identified as non-normal.

Table 2: Characteristics for participants with diabetes (male vs. female)

	Males (n = 1251)		**Females (n = 950)**		**p-Value**
N = 2201	***Mean (SD)***	***Median (IQR)***	***Mean (SD)***	***Median (IQR)***	
Age (years)	66 (9)	66 (59-73)	64 (10)	64 (57-72)	.006[a]
Age of Diagnosis (years) [c]	57 (13)	57 (50-65)	54 (16)	56 (46-65)	.002[b]
Distensibility ($mmHg^{-1}$)	.0021 (.0010)	.0020 (.0014-.0027)	.0022 (.0010)	.0020(.0014-.0027)	.972[b]
cIMT (mm)	.77 (.18)	.75 (.65-.88)	.74 (.16)	.73 (.63-.83)	<.001[a]
BMI [c]	29.1 (4.6)	28.8 (25.9-31.6)	29.9 (6.0)	29.4 (25.4-33.6)	<.001[a]
hs-CRP (mg/L)	2.3 (4.6)	1.1 (.7-2.4)	3.4 (6.1)	1.8 (.9-3.7)	<.001[b]
HbA1c (%)	6.6 (1.2)	6.2 (5.7-7.0)	6.3 (1.1)	5.9 (5.6-6.6)	<.001[b]
Triglycerides (mmol/L)	2.02 (1.17)	1.69 (1.20-2.55)	1.95 (1.03)	1.72 (1.24-2.39)	.985 [b]
HDL (mM)	1.23 (.39)	1.16 (.96-1.44)	1.52 (.45)	1.43 (1.19-1.76)	<.001[b]
LDL (mM) [c]	2.24 (.94)	2.06 (1.54-2.87)	2.63 (1.02)	2.55 (1.87-3.33)	<.001[a]
Cholesterol (mM)	4.36 (1.09)	4.21 (3.54-5.05)	5.03 (1.18)	4.97 (4.17-5.78)	<.001 [a]
TC/HDL Ratio	3.82 (1.39)	3.55 (2.80-4.50)	3.56 (1.24)	3.33 (2.70-4.08)	<.001[a]
SBP (mmHg)	124.1 (16.0)	122.4 (113.2-133.6)	122.3 (16.4)	121.0 (111.2-131.6)	.009[a]
DBP (mmHg)	74.9 (10.1)	74.6 (68.0-81.2)	71.4 (9.6)	71.6 (64.6-77.9)	<.001[a]
Maximal Grip Strength (kg) [c]	41.40 (9.46)	41.13 (35.18-47.32)	26.07(6.13)	25.57 (21.99-29.80)	<.001[a]
PASE Score [c]	138.9 (70.6)	127.3 (88.6-179.6)	121.4 (66.4)	111.00 (72.1-159.5)	<.001[b]
Get Up and Go Time (seconds) [c]	10.03 (2.32)	9.67 (8.56-10.98)	10.05 (2.48)	9.60 (8.49-11.16)	.707[b]
4 Metre Walk Time (seconds) [c]	4.37 (1.01)	4.19 (3.71-4.81)	4.49 (1.09)	4.28 (3.75-5.04)	.005[b]
Total Chair Rise Time (seconds) [c]	13.76 (3.60)	13.37 (11.3-15.62)	14.02 (3.9)	13.52 (11.47-16.09)	.267[b]

p-Values <.050 were considered statistically significant. [a] p-Value determined by an independent samples t-test, males vs. females; [b] p-value determined by a Mann-Whitney U test; [c] Variable was missing for less than 9% of participants.

Table 3: Characteristics for participants with T1DM vs. participants with DM2O

	T1DM (n = 75)		DM2O (n = 2126)		p-Value
N = 2201	***Mean (SD)***	***Median (IQR)***	***Mean (SD)***	***Median (IQR)***	
Age (years)	62 (10)	62 (54-70)	65 (10)	65 (58-72)	.016[a]
Age of Diagnosis [c] (years)	35 (19)	30 (18-51)	56 (13)	57 (50-65)	<.001[b]
Distensibility ($mmHg^{-1}$)	.0025 (.0011)	.0019 (.0016-.0027)	.0021 (.0010)	.0020 (.0014-.0027)	.792[b]
cIMT (mm)	.75 (.16)	.72 (.63-.86)	.76 (.17)	.74 (.64-.86)	.755[a]
BMI [c]	27.2 (4.9)	26.5 (23.3-29.8)	29.6 (5.2)	29.0 (25.8-32.4)	<.001[a]
hs-CRP (mg/L)	3.1 (6.3)	1.0 (.7-3.0)	2.8 (5.3)	1.3 (.7-2.9)	.476[b]
HbA1c (%)	7.7 (1.2)	7.5 (6.8-8.5)	6.4 (1.2)	6.0 (5.6-6.8)	<.001[b]
Triglycerides (mM)	1.48 (1.01)	1.16 (.87-1.69)	2.01 (1.11)	1.72 (1.24-2.47)	<.001[b]
HDL (mM)	1.61 (.53)	1.60 (1.16-1.94)	1.34 (.44)	1.27 (1.03-1.57)	<.001[b]
LDL (mM) [c]	2.01 (.72)	1.85 (1.62-2.41)	2.43 (1.00)	2.28 (1.66-3.10)	<.001[a]
Cholesterol (mM)	4.27 (.82)	4.19 (3.63-4.75)	4.66 (1.19)	4.52 (3.76-5.45)	<.001[a]
TC/HDL Ratio	2.91 (1.04)	2.64 (2.18-3.32)	3.73 (1.33)	3.47 (2.78-4.33)	<.001[b]
SBP (mmHg)	121.9 (16.7)	118.8 (108.2-135.0)	123.4 (16.2)	121.8 (112.6-132.6)	.421[a]
DBP (mmHg)	69.7 (10.0)	68.2 (62.8-75.6)	73.6 (10.0)	73.6 (66.4-80.0)	<.001[a]
Maximal Grip Strength (kg) [c]	31.59 (9.38)	29.94 (24.95-38.22)	35.04 (11.22)	34.06 (26.04-42.95)	.005[a]
PASE Score [c]	134.3 (79.6)	120.3 (67.4-190.2)	131.3 (69.0)	119.6 (82.8-171.0)	.960[b]
Get Up and Go Time (seconds) [c]	10.12 (2.44)	9.74 (8.79-11.13)	10.04 (2.39)	9.63 (8.53-11.03)	.562[b]
4 Metre Walk Time (seconds) [c]	4.47 (.95)	4.40 (3.93-4.88)	4.42 (1.05)	4.22 (3.72-4.91)	.674[a]
Total Chair Rise Time (seconds) [c]	13.69 (3.31)	13.31 (11.42-15.47)	13.88 (3.74)	13.41 (11.37-15.82)	.671[a]

p-Values <0.050 were considered statistically significant. [a] p-Value determined by an independent samples t-test, type 1 vs. other; [b] p-value determined by a Mann-Whitney U test; [c] Variable was missing for less than 9Ind% of participants.

Table 4: Sample breakdown by diabetes type and sex

	T1DM			DM2O		
	Sample n	***Sample %***	***Total Cohort % [a]***	***Sample n***	***Sample %***	***Total Cohort % [a]***
Total	75	3.4%	0.3%	2126	96.6%	7.1%
Male	38	1.7%	0.1%	1213	55.1%	4.0%
Female	37	1.7%	0.1%	913	41.5%	3.0%

[a] Total cohort refers to the total CLSA Comprehensive data set (N= 30,097).

Table 5: Sample breakdown for Hypertension Data

N = 2,201	**% of Sample with Hypertension**		**% of Sample taking medication for Hypertension**	
	n	***%***	***n***	***%***
Total	1,222	55.6%	1035	47.0%
Type 1	39	1.8%	37	1.7%
DM2O	1183	53.7%	998	45.3%
Male	704	31.99%	635	28.9%
Type 1	19	0.9%	15	0.7%
DM2O	685	31.2%	620	28.2%
Female	518	23.5%	400	18.2%
Type 1	20	0.9%	22	0.99%
DM2O	498	22.6%	378	17.2%

Percentages are expressed relative to the total sample of 2,201.

Table 6: Sample Breakdown for Self-Reported Heart Disease Data

N = 2,201	**% of Sample with Heart Disease**	
	n	***%***
Total	381	17.3%
Type 1	18	.8%
DM2O	363	16.5%
Male	215	9.8%
Type 1	9	.4%
DM2O	206	9.4%
Female	166	7.5%
Type 1	9	0.4%
DM2O	157	7.1%

Percentages are expressed relative to the total sample of 2,201.

Table 7: Spearman's Rho Correlation Matrix

Variable	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22
1.Distensibility (mm)	1																					
2. Sex	.001	1																				
3. Age (years)	-.317**	.061*	1																			
4. Age of Diagnosis (years) (n=2190)	-.199**	.065**	.696**	1																		
5. cIMT (mm) (n=2015)	-.109**	.084**	.398**	.257**	1																	
6. BMI (n=2197)	-.117**	-.054**	-.125**	-.047**	.063**	1																
7. hs-CRP (mg/L)	-.087**	-.185**	-.039*	-.011	.012	.410**	1															
8. HbA_{1c} (%)	-.176**	.137**	.153**	.030	.128**	.174**	.132**	1														
9.Triglycerides (mmol/L)	-.138**	-.000	-.082**	-.000	-.006	.298**	.207**	.184**	1													
10. HDL (mmol/L)	.085**	-.338**	.067**	.021	-.060*	-.320**	-.161**	-.265**	-.550**	1												
11. LDL (mmol/L) (n=2122)	.079**	-.200**	-.222**	-.091**	-.078**	-.053**	.113**	-.334**	.061**	.191**	1											
12.Cholesterol (mmol/L)	.047*	-.288**	-.203**	-.081**	-.088**	-.067**	.106**	-.301**	.210**	.313**	.924**	1										
13. SBP (mmHg)	-.364**	.054*	.179**	.130**	.151**	.197**	.143**	.105**	.094**	-.047*	.017	.034	1									
14. DBP (mmHg)	-.119**	.165**	-.314**	-.140**	-.118**	.206**	.115**	-.027	.141**	-.112**	.200**	.178**	.553**	1								
15. TC/HDL Ratio	-.048*	.096**	-.206**	-.070**	-.008**	.264**	.243**	.018	.685**	-.678**	.535*	.438**	.074**	.240**	1							
16. Max Grip Strength (kg) (n=2053)	.096**	.711**	-.255**	-.115**	-.035	.040	-.163**	-.016	.019	-.247**	-.014	-.085**	.038	.274**	.165**	1						
17. PASE Score (n=2074)	.151**	.129**	-.335**	-.185**	-.137**	-.026	-.116**	-.138**	-.056*	.021	.130**	.090**	-.055*	.149**	-.048*	.299**	1					
18. Diagnosed with Hypertension (n=2195)	.148**	-.019	-.201**	-.135**	-.126**	-.183**	-.098**	-.125**	-.082**	.090**	.196**	.177**	-.256**	-.039	.029	.082**	.142**	1				
19. Current Tx for Hypertension (n=2149)	-.096**	.090**	.219**	.143**	.102**	.114**	.034	.198**	.023	-.098**	-.310**	-.297**	.061**	-.088**	-.123**	-.037	-.123**	-.423**	1			
20. Get Up and Go Time (seconds) (n=2185)	-.160**	.008	.352**	.205**	.179**	.165**	.146**	.178**	.045*	-.097**	-.144**	-.137**	.118**	-.118**	-.010	-.243**	-.253**	-.169**	.149**	1		
21. 4- Metre Walk Test Time (seconds) (n=2191)	-.166**	-.060**	.318**	.183**	.161**	.134**	.148**	.166**	.055*	-.063**	-.125**	-.111**	.095**	-.144**	-.017	-.297**	-.262**	-.135**	.118**	.740**	1	
22. Total Chair Rise Test Time (seconds) (n=2092)	-.092**	-.024	.193**	.118**	.080**	.077**	.077**	.127**	.038	-.078**	-.095**	-.091**	.087**	-.047*	.004	-.185**	-.164**	-.102**	.051**	.521**	.373**	1

** Correlation is significant at the .010 level (2-tailed). * Correlation is significant at the .050 level (2-tailed).

Table 8: Initial Unadjusted Model for PASE and Distensibility

Distensibility	B	T	Sig.	R^2	ΔR^2
Model				.086	.084
Intercept	3.911E-3	17.959	<.001		
Age	-2.683E-5	-8.196	<.001		
Sex	5.89E-4	1.799	.072		
PASE score	1.882E-6	.848	.397		
Age*sex	-9.532E-6	-1.904	.057		
Age*PASE	-2.815E-8	-.807	.420		
Sex*PASE	-1.088E-7	-.152	.879		

P-value significant at the .050 level.

Table 9: Unadjusted Model (outliers and interactions removed)

Distensibility	B	T	Sig.	R^2	ΔR^2
Model				.107	.106
Intercept	4.068E-3	27.871	<.001		
Age	-2.990E-5	-13.472	<.001		
Sex	-2.679E-5	-.674	.500		
PASE score	4.457E-7	1.466	.143		

P-value significant at the .050 level.

Table 10: Adjusted Model

Distensibility	B	T	Sig.	R^2	ΔR^2
Model				.248	.242
Intercept	4.233E-3	31.511	<.001		
Age	-3.333E-5	-12.317	<.001		
Sex	-9.370E-5	-2.060	.040		
PASE	7.331E-10	.002	.998		
cIMT	3.070E-4	2.510	.012		
HBA_{1C}	-8.063E-5	-4.286	<.001		
hsCRP	-1.907E-6	-.532	.595		
Triglycerides	-9.396E-5	-3.585	<.001		
HDL-C	2.367E-6	-.041	.967		
LDL-C	2.850E-5	1.318	.188		
SBP	-1.321E-5	-7.770	<.001		
DBP	-6.857E-6	-2.385	.017		
BMI	-1.406E-5	-3.382	<.001		
Self-reported heart disease	-9.765E-5	-1.911	.056		
Smoker/previous smoker	1.170E-4	1.716	.086		

P-value significant at the .050 level.

2.4 Discussion

In the current study, we sought to examine the influence of PA on carotid artery stiffness as measured in the baseline data sets from a large cohort of older Canadians with DM classified as DM2O after controlling for known CVD risk factors and markers, and whether any differences across age and sex existed. The main findings of this project are two-fold: (1) physical activity as assessed by the PASE is not associated with arterial distensibility in older adults with DM2O, and (2) these results did not differ based on age or sex.

It was hypothesized that PASE would be associated with accelerated carotid arterial distensibility, a direct measure of arterial stiffness, in older adults with DM2O. This hypothesis was grounded on a breadth of literature supporting the notion that higher levels of habitual physical activity and physical fitness are associated with attenuated age-related arterial stiffening in healthy older adults [12,13,117–121]. The present study found that PASE was not associated with arterial distensibility in older adults with DM2O. While PASE was significantly correlated with arterial distensibility, there was no main effect of PASE on arterial distensibility before and after adjustment for known CVD risk factors and markers.

The findings of the present study do not necessarily suggest that increased physical activity is unrelated to favourable outcomes regarding improved vascular health. Instead, it is important to consider that the indices with which both arterial stiffness and physical activity were assessed differ from much of the previously cited literature [117,122]. Tanaka *et al.* (1998) demonstrated attenuated age-related arterial stiffening via aortic pulse-wave velocity (PWV) in post-menopausal females. Similar findings were corroborated by Kozakova *et al.* (2007), who assessed carotid-femoral (cfPWV) and objectively measured PA in middle-aged males and females and found attenuations in age-related vascular stiffening in more physically active

individuals. Carotid arterial distensibility measures local arterial stiffness of an elastic artery, while cfPWV reflects the properties of multiple segments, including elastic and muscular arteries [89]. As such, it is possible that a greater representation of the arterial tree including more than just the central arterial regions is necessary to detect changes due to PA. Indeed, higher levels of moderate-vigorous PA were associated with lower levels of baPWV, a composite metric of arterial stiffness, in middle-aged adults with obesity [126].

While Königstein *et al.* (2020) recently demonstrated that arterial stiffness was strongly associated with objectively measured MVPA in middle-aged adults with obesity, Stamatelopoulos *et al.* (2020) recently found that subjectively measured PA was inversely associated with arterial stiffness in normal weight but not obese, post-menopausal women[126,127]. Several methodological differences exist between these two studies. Königstein *et al.* (2020) assessed baPWV, which indicates peripheral arterial stiffness, while Stamatelopoulos et al. (2020) assessed cfPWV, which indicates central arterial stiffness. It is known that central arterial stiffness is more strongly associated with of CVD risk compared to peripheral arterial stiffness [138]. Furthermore, PA was assessed differently between studies (objective vs. subjective assessment of PA). Finally, differences in sample characteristics may account for the discrepant findings, as Stamatelopoulos *et al.* (2020) examined women only and an older age group. This discrepancy highlights the need to examine the influence of subjectively measured PA on central arterial stiffness in an older group of adults with an elevated cardiometabolic risk, including both biological sexes.

Like the present study, Schmitz *et al.* (2002) assessed arterial distensibility and subjectively measured PA in older males and females but found only a weak association between work-related PA and arterial distensibility. It was suggested that habitual PA might not be

associated with arterial stiffness [118]. In contrast, Tanaka *et al.* (2018) found higher PA in later life, and habitual PA from mid- to later life was associated with lower central arterial stiffness. Despite minor attenuation, these findings persisted after adjusting for diabetes and hypertension [12]. A strength of the study from Tanaka *et al.* (2018) used the same Baecke questionnaire in a repeated measures design, which allowed for the assessment of long term (6- year follow-up) habitual PA. Schmitz *et al.* (2001) presented PA data using a modified Baecke questionnaire, which accounted for PA of different intensities over the last 10 months [123]. The PASE used in the present study accounts for PA performed over 7 days [132]. Both studies using subjective assessments are cross-sectional, and thus causality cannot be inferred. As such, the results of Tanaka *et al.* (2018), most notably the finding that mid-late life high PA was associated with lower arterial stiffness even after adjustment for diabetes, are promising and highlight the need to perform follow-up analysis on PASE and carotid arterial distensibility in this group once these data become available.

The PASE is designed to assess PA habits in older adults aged 65 and older, accounting for PA typically performed by older persons [132]. The mean age of the present study was approximately 65 years, with a range of 45-85 years. One thousand seventy individuals were between the ages of 45 and 64. As such, it is possible that the PASE was not the most appropriate assessment of PA in a portion of this population for a cross-sectional analysis of baseline data. However, the present study is part of a larger, longitudinal study on aging [128]. As such, PASE was selected to assess PA throughout aging, as many individuals included in the baseline study will eventually be over the age of 65, at which point PASE will be an appropriate metric. Furthermore, it may be postulated that PASE scores were simply too low and tightly scattered about the mean to detect any linear relationship. However, Logan *et al.* (2013)

suggested *via* correlational data that a minimum PASE score of ~140 for males and ~120 for females was associated with a favourable waist circumference, a known predictor of arterial stiffness in middle-older adults [115,139]. The mean PASE scores in the present study were approximately 139 in males and 121 in females, nearly equivalent to the recommendations of Logan *et al.* (2013), thus suggestive that PASE scores were sufficient to result in favourable health outcomes related to arterial stiffness. Finally, the PASE is a subjective assessment of PA. It is unlikely that the lack of relationship between PASE and arterial distensibility in the present study is limited by the PASE scale and more likely limited by the cross-sectional nature of the present study.

Duration of diabetes was not considered in the present study, but has been shown to be associated with arterial stiffness in older adults with T2DM [140]. Diabetic complications are known to increase with a greater duration of disease, and the risk of coronary heart disease was 1.38 times higher and risk of CHD death was 1.8 times higher for each decade of diabetes duration [140,141]. In individuals with a shorter disease duration (0.3-6.2 years), increased risk of mortality was attributed to other CVD risk factors, such as prior myocardial infarction and microalbuminuria. However, in individuals with a longer duration of diabetes (6.3-29.0 years), the elevated risk of mortality was almost completely attributed to the length of disease, independent of other CVD factors [142]. As such, follow up analysis should include diabetes duration as an independent risk factor.

It was hypothesized that sex differences would modulate any observed relationships between PA and arterial distensibility. While we did not observe any main effect of sex in the initial model that included only age, sex and PASE (P=.536), there was a main effect for sex that approached insignificance and should be interpreted with caution (P=.040) when adjusting for

age, PASE, heart disease, smoking status, hsCRP, HbA_{1c}, SBP, DBP, cIMT, Triglycerides, HDL-C, and LDL-C. In our baseline characteristics comparisons, arterial distensibility did not differ significantly between males and females (*P*=.972). It is known that sex differences exist in older adults with regards to arterial stiffness, such that increases in arterial stiffness are typically greater in older females compared to older males as assessed by both PWV [44] and carotid arterial distensibility [143]. The greater arterial stiffness in older women is typically attributed to menopause and the resulting loss of the cardioprotective effects of estrogen [19]. In a cohort of middle-aged adults with, and without, T2DM, the age-related increase in arterial stiffness was more pronounced in females with T2DM than females without T2DM, and no difference was found in males [144]. The current findings regarding a lack of sex differences in the relationships between arterial stiffness and PA in DM2O do not necessarily refute previous findings in this area; however, they highlight a limitation of the cross-sectional nature of the present study. Longitudinal analysis will likely be required to detect any sex differences in the progression of arterial stiffness over time.

It is important to consider the characteristics of the sample with self-reported hypertension and heart disease. It was found that 1,183 (56%) of the 2,126 individuals in the DM2O cohort self-reported a diagnosis of hypertension, and 998 (47%) individuals in the DM2O cohort self-reported that they were currently taking anti-hypertensive medication. The fact that over 50% of this sample reported a diagnosis of hypertension is unsurprising, as it has been suggested that stiffening of the arterial wall can a precursor to the development of hypertension [145]. Furthermore, 363 (17%) of the 2126 self-reported a diagnosis of heart disease, which is also unsurprising in the aging and diabetic population in the present study, as diabetes and aging are both implicated in the progression of arterial stiffness. With aging, arterial walls thicken and lose

elasticity, resulting in impaired vascular function, especially in the central arteries located proximally to the heart.[6], and this process is accelerated in the diabetic state [7–11].

A weak negative correlation between the serum inflammatory marker hsCRP and arterial distensibility was observed, in the present study. However, no main effect of hsCRP on arterial distensibility was found in the adjusted general linear model. hsCRP is involved in the synthesis of tissue factors, cytokines, and platelet aggregation, and as such, it is heavily involved in the atherosclerotic development process. This inflammatory marker is elevated in acute and chronic inflammation, peripheral vascular diseases, diabetes, and hypertension [86]. In a large sample of adults with diabetes (n = 1614; age 17-66+), it was found that increased HbA_{1c} was associated with higher levels of hsCRP, and thus chronic inflammation [79], which is consistent with the present study. This relationship suggests accelerated vascular stiffness in diabetic populations [78]. In a group of 362 middle-aged and elderly males (mean age 60 ± 11 years), Nakhai-Pour *et al.* (2007) assessed aortic PWV and hsCRP. It was found that individuals with diabetes mellitus who were older had higher levels of hsCRP. Furthermore, it was found that hsCRP was predictive of PWV and thus arterial stiffness, such that aortic PWV increased significantly with higher levels of hsCRP [78], a finding that is inconsistent with the present study, and results regarding carotid arterial distensibility and hsCRP in adults with diabetes of any type are extremely limited, highlighting the need for further research in this area.

2.4.1 Strengths and Limitations

Several strengths of the present study exist that make the findings of this thesis a valuable contribution to the literature in this area. Using a national longitudinal database (CLSA) [128] allowed for the inclusion of a sample size (n=2,201) that would have been otherwise unfeasible. Regarding the analysis of sex differences, this study included a sample of both males (57% of the

sample) and females (43% of the sample) and was therefore not limited by including both men and women. Furthermore, the use of the CLSA database, with follow-up collection is planned for every 3 years for 20 years [128], should be considered a strength. In the future, this will provide several follow-up points, allowing for comparative analysis with the present findings and the detection of longitudinal, within-subject comparisons, and potential sex-based differences.

The present study also used empirical diagnostic criteria [5] to discriminate individuals with T1DM from the DM2O individuals. Differences exist in both the pathophysiology and manifestation of the two types of diabetes, and it is important to distinguish between them in many investigations[5]. Individuals with T1DM are typically not overweight, while over 90% of individuals with T2DM are at least overweight, if not obese [5]. As such, the associated risk factors and markers of the two diseases can differ, as noted in the differing BMI, HbA_{1c}, triglyceride, and cholesterol variables observed between disease groups in the present study (Table 3). As such, the isolated analysis of DM2O is a strength of the present study. Furthermore, a full profile of cardiovascular risk factors and markers typically associated with diabetes was included in the present study. With access to the cardiovascular disease risk profile, the findings of the present study regarding the lack of relationship between PASE and arterial distensibility in this population are likely independent of the influence of other common risk factors and markers. Use of a weighted score, such as the INTERHEART Risk Score[146] , may have allowed for a more appropriate determination of the effect of several CVD risk factors and markers. The INTERHEART Risk Score is a valid composite CVD risk score that includes age, sex, smoking status, diabetes, high blood pressure, familial history of heart disease, waist-to-hip ratio, psychosocial factors, diet, and physical activity. Greater scores indicate a greater CVD risk [147].

Several limitations of the present study cannot be overlooked. In the CLSA, BP was assessed at the brachial artery before ultrasound imaging with the participant in a seated position, while the ultrasound images used to calculate arterial distensibility were obtained at the carotid artery with the participant in the supine position. In assessing arterial distensibility, blood pressure can be assessed locally via applanation tonometry on the carotid artery opposite to the one being imaged, or peripherally, using a standard brachial blood pressure cuff [32,54]. It is typically recommended that BP as an independent metric be assessed in a seated position, as supine BP measurements can be lower on average [148]. Local BP values can differ from brachial BP values due to pulse amplification [32]. BP is required to calculate pulse pressure, which is used in the calculation of arterial distensibility [51]. Arterial distensibility is itself a measurement of local arterial stiffness within a certain region, and the change in arterial diameter depends on the change in arterial pressure [32]. As such, local assessment of BP is typically recommended when assessing arterial distensibility; however, this study may be limited by equipment availability, and tonometry can be difficult to assess in certain clinical populations, such as DM [32,55].

This study is also limited by the inability to distinguish between Type 2 and "other" diabetes. Diabetes type was assessed via self-report during baseline assessment. T2DM accounts for 90-95% of diabetes cases, and this is consistent with the present study, in which "other" diabetes accounted for almost 97% of cases. The DM2O category may also include females who previously had gestational diabetes, which can occur during pregnancy [5]. However, the mean age of diagnosis was 56 years in the present study, and BMI was significantly greater in the DM2O category (29.6 vs. 27.2 P= <.001). These findings would place the mean BMI in the other category as overweight/obese, as a BMI between 25.0 to <30 kg/m^2 is considered overweight [113]. A BMI of 30.0 kg/m^2 or higher is considered obese [113]. Punthakee *et al.* (2018) reported that the

mean age of diagnosis is typically older in T2DM (>25 years) compared to T1DM, and roughly 90% of T2DM cases are overweight. Furthermore, the mean HbA_{1c} in the DM2O group was 6.4%, which is considered pre-diabetic [5]. However, HbA_{1c} is an indirect measure of average blood glucose levels, and other factors should be considered that may impact hemoglobin glycation independently of glycemia, including age, race/ ethnicity, pregnancy status, genetic background, and anemia/hemoglobinopathies [59]. As such, further criteria would be required to properly ascertain Type 2 diabetics from the rest of the sample, such as fasting plasma glucose (FPG), 2-hour (2-h) post-load plasma glucose after a 75 g oral glucose tolerance test (2hPG in a 75 g OGTT), or a random blood glucose test [56].

2.4.2 Future Directions

As further data is collected in the CLSA sample, longitudinal measures of carotid artery distensibility will become. available. Future research should examine both changes in PA over time, as well between group comparisons. It was previously mentioned that the cross-sectional nature of the current study might have limited interpretations of PA data and repeat measures of both PA and distensibility would allow for the detection of a relationship in either direction. Furthermore, the present study did not seek to compare diabetes vs. non-diabetes. Future research should examine the cross-sectional baseline data to compare relationships between PASE and distensibility in diabetics vs. non-diabetics, which would allow for mechanistic insight regarding the metabolic syndrome and arterial distensibility.

2.5 Conclusion

The results of this study suggest that, despite a weak negative correlation, PA, as measured by the PASE tool was not associated with arterial distensibility in older adults with DM2O before and after adjustment for known CVD risk factors and markers. These results did

not differ based on age or sex, and no sex differences were found for arterial distensibility in this population. Follow-up analysis is recommended to assess the influence of long-term PA on arterial distensibility and whether any progression rather than the absolute values of arterial stiffness differ based on sex in this population.

References

1. World Health Organization. *Cardiovascular Diseases (CVDs)*.; 2017.
2. Virani SS, Alonso A, Aparicio HJ, et al. Heart Disease and Stroke Statistics-2021 Update: A Report From the American Heart Association. *Circulation*. 2021;143(8). doi:10.1161/CIR.0000000000000950
3. Yazdanyar A, Newman AB. The Burden of Cardiovascular Disease in the Elderly: Morbidity, Mortality, and Costs. *Clin Geriatr Med*. 2009;25(4). doi:10.1016/j.cger.2009.07.007
4. Rodgers JL, Jones J, Bolleddu SI, et al. Cardiovascular Risks Associated with Gender and Aging. *J Cardiovasc Dev Dis*. 2019;6(2). doi:10.3390/jcdd6020019
5. Punthakee Z, Goldenberg R, Katz P. Definition, Classification and Diagnosis of Diabetes, Prediabetes and Metabolic Syndrome. *Can J Diabetes*. 2018;42:S10-S15. doi:10.1016/j.jcjd.2017.10.003
6. Sun Z. Aging, arterial stiffness, and hypertension. *Hypertension*. 2015;65(2):252-256. doi:10.1161/HYPERTENSIONAHA.114.03617
7. Atabek ME, Kurtoglu S, Pirgon O, Baykara M. Arterial wall thickening and stiffening in children and adolescents with type 1 diabetes. *Diabetes Res Clin Pract*. 2006;74(1). doi:10.1016/j.diabres.2006.03.004
8. Giannattasio C, Failla M, Piperno A, et al. Early impairment of large artery structure and functionin Type I diabetes mellitus. *Diabetologia*. 1999;42:987-994.
9. Llauradó G, Ceperuelo-Mallafré V, Vilardell C, et al. Arterial stiffness is increased in patients with type 1 diabetes without cardiovascular disease: A potential role of low-grade inflammation. *Diabetes Care*. 2012;35(5):1083-1089. doi:10.2337/dc11-1475
10. Rydén Ahlgren A˚, Sundkvist G, Sandgren T, Länne T, Ahlgren R. *Female Gender Increases Stiffness of Elastic but Not of Muscular Arteries in Type I Diabetic Patients Accepted for Publication*.
11. Kimoto E, Shoji T, Shinohara K, et al. Preferential stiffening of central over peripheral arteries in type 2 diabetes. *Diabetes*. 2003;52(2):448-452. doi:10.2337/diabetes.52.2.448
12. Tanaka H, Palta P, Folsom AR, et al. Habitual Physical Activity and Central Artery Stiffening in Older Adults: The ARIC Study. *J Hypertens*. 2018;36(9):1889-1894. doi:10.1097/HJH.0000000000001782
13. Kozakova M, Palombo C, Mhamdi L, et al. Habitual physical activity and vascular aging in a young to middle-age population at low cardiovascular risk. *Stroke*. Published online 2007. doi:10.1161/STROKEAHA.107.484949
14. Kakiyama T, Matsuda M, Koseki S. Effect of physical activity on the distensibility of the aortic wall in healthy males. *Angiology*. 1998;49(9). doi:10.1177/000331979804901007
15. Peterson MJ, Giuliani C, Morey MC, et al. Physical activity as a preventative factor for frailty: The health, aging, and body composition study. *Journals Gerontol - Ser A Biol Sci Med Sci*. 2009;64(1). doi:10.1093/gerona/gln001

16. Kasapis C, Thompson PD. The effects of physical activity on serum C-reactive protein and inflammatory markers: A systematic review. *J Am Coll Cardiol*. Published online 2005. doi:10.1016/j.jacc.2004.12.077

17. Oh YS. Arterial stiffness and hypertension. *Clin Hypertens*. 2018;24(17).

18. Mercadante AA, Raja A. *Anatomy, Arteries. [Updated 2021 Jan 13]. In: StatPearls [Internet]*. StatPearls Publishing; 2021. Accessed May 25, 2021. https://www.ncbi.nlm.nih.gov/books/NBK547743/?report=reader#_NBK547743_pubdet_

19. DuPont JJ, Kenney RM, Patel AR, Jaffe IZ. Sex differences in mechanisms of arterial stiffness. *Br J Pharmacol*. 2019;176(21):4208-4225. doi:10.1111/bph.14624

20. Majesky MW, Dong XR, Hoglund V, Mahoney WM, Daum G. The adventitia: A dynamic interface containing resident progenitor cells. *Arterioscler Thromb Vasc Biol*. 2011;31(7). doi:10.1161/ATVBAHA.110.221549

21. Nichols WW, Denardo SJ, Wilkinson IB, McEniery CM, Cockcroft J, O'Rourke MF. Effects of arterial stiffness, pulse wave velocity, and wave reflections on the central aortic pressure waveform. *J Clin Hypertens*. 2008;10(4). doi:10.1111/j.1751-7176.2008.04746.x

22. Cecelja M, Chowienczyk P. Role of arterial stiffness in cardiovascular disease. *JRSM Cardiovasc Dis*. Published online 2012. doi:10.1258/cvd.2012.012016

23. Avolio A. Arterial Stiffness. *Pulse*. 2013;1(1):14-28. doi:10.1159/000348620

24. Zieman SJ, Melenovsky V, Kass DA. Mechanisms, pathophysiology, and therapy of arterial stiffness. *Arterioscler Thromb Vasc Biol*. 2005;25(5). doi:10.1161/01.ATV.0000160548.78317.29

25. Xu C, Zarins CK, Pannaraj PS, Bassiouny HS, Glagov S. Hypercholesterolemia superimposed by experimental hypertension induces differential distribution of collagen and elastin. *Arterioscler Thromb Vasc Biol*. 2000;20(12). doi:10.1161/01.ATV.20.12.2566

26. Johnson CP, Baugh R, Wilson CA, Burns J. Age related changes in the tunica media of the vertebral artery: Implications for the assessment of vessels injured by trauma. *J Clin Pathol*. 2001;54(2). doi:10.1136/jcp.54.2.139

27. Harper S. Economic and social implications of aging societies. *Science (80-)*. 2014;346(6209). doi:10.1126/science.1254405

28. Benetos A, Waeber B, Izzo J, et al. Influence of age, risk factors, and cardiovascular and renal disease on arterial stiffness: Clinical applications. *Am J Hypertens*. 2002;15(12). doi:10.1016/S0895-7061(02)03029-7

29. Haluska BA, Jeffries L, Carlier S, Marwick TH. Measurement of arterial distensibility and compliance to assess prognosis. *Atherosclerosis*. 2010;209(2). doi:10.1016/j.atherosclerosis.2009.10.018

30. Mitchell GF, Hwang SJ, Vasan RS, et al. Arterial stiffness and cardiovascular events: The framingham heart study. *Circulation*. Published online 2010. doi:10.1161/CIRCULATIONAHA.109.886655

31. Lakatta EG, Levy D. Arterial and Cardiac Aging: Major Shareholders in Cardiovascular Disease Enterprises Part I: Aging Arteries: A "Set Up" for Vascular Disease The

Demographic Imperative and the Risk of Vascular Diseases in Older Persons. Published online 2003. doi:10.1161/01.CIR.0000048892.83521.58

32. Segers P, Rietzschel ER, Chirinos JA. How to Measure Arterial Stiffness in Humans. *Arterioscler Thromb Vasc Biol*. Published online 2020. doi:10.1161/ATVBAHA.119.313132

33. Bia D, Aguirre I, Zócalo Y, Devera L, Cabrera Fischer E, Armentano R. Regional Differences in Viscosity, Elasticity, and Wall Buffering Function in Systemic Arteries: Pulse Wave Analysis of the Arterial Pressure-Diameter Relationship. *Rev Española Cardiol (English Ed*. 2005;58(2). doi:10.1016/s1885-5857(06)60360-5

34. Wang M, Jiang L, Monticone RE, Lakatta EG. Proinflammation: The key to arterial aging. *Trends Endocrinol Metab*. 2014;25(2). doi:10.1016/j.tem.2013.10.002

35. Deanfield JE, Halcox JP, Rabelink TJ. Endothelial function and dysfunction: Testing and clinical relevance. *Circulation*. Published online 2007:1285-1295. doi:10.1161/CIRCULATIONAHA.106.652859

36. Calles-Escandon J, Cipolla M. Diabetes and endothelial dysfunction: A clinical perspective. *Endocr Rev*. Published online 2001. doi:10.1210/edrv.22.1.0417

37. Quyyumi AA. Endothelial function in health and disease: New insights into the genesis of cardiovascular disease. In: *American Journal of Medicine*. ; 1998. doi:10.1016/s0002-9343(98)00209-5

38. Celermajer DS, Sorensen KE, Spiegelhalter DJ, Georgakopoulos D, Robinson J, Deanfield JE. Aging is associated with endothelial dysfunction in healthy men years before the age-related decline in women. *J Am Coll Cardiol*. 1994;24(2). doi:10.1016/0735-1097(94)90305-0

39. LaRocca TJ, Martens CR, Seals DR. Nutrition and other lifestyle influences on arterial aging. *Ageing Res Rev*. 2017;39. doi:10.1016/j.arr.2016.09.002

40. Ren L, Cai J, Liang J, Li W, Sun H. Impact of cardiovascular risk factors on carotid intima-media thickness and degree of severity: A cross-sectional study. *PLoS One*. Published online 2015. doi:10.1371/journal.pone.0144182

41. Strawbridge RJ, Ward J, Bailey MES, et al. Carotid intima-media thickness novel loci, sex-specific effects, and genetic correlations with obesity and glucometabolic traits in UK Biobank. *Arterioscler Thromb Vasc Biol*. 2020;(February):446-461. doi:10.1161/ATVBAHA.119.313226

42. Tanaka H, Dinenno FA, Monahan KD, DeSouza CA, Seals DR. Carotid artery wall hypertrophy with age is related to local systolic blood pressure in healthy men. *Arterioscler Thromb Vasc Biol*. 2001;21(1):82-87. doi:10.1161/01.ATV.21.1.82

43. Gómez-Marcos MT, Recio-Rodríguez JI, Patino-Alonso MC, et al. Relationship between intima-media thickness of the common carotid artery and arterial stiffness in subjects with and without type 2 diabetes: A case-series report. *Cardiovasc Diabetol*. 2011;10:1-8. doi:10.1186/1475-2840-10-3

44. Coutinho T. Arterial stiffness and its clinical implications in women. *Can J Cardiol*. Published online 2014. doi:10.1016/j.cjca.2014.03.020

45. Zaydun G, Tomiyama H, Hashimoto H, et al. Menopause is an independent factor augmenting the age-related increase in arterial stiffness in the early postmenopausal phase. *Atherosclerosis*. Published online 2006. doi:10.1016/j.atherosclerosis.2005.03.043

46. Hildreth KL, Kohrt WM, Moreau KL. Oxidative stress contributes to large elastic arterial stiffening across the stages of the menopausal transition. *Menopause*. Published online 2014. doi:10.1097/GME.0000000000000116

47. Sztejnsznajd C, da Silva MER, Nussbacher A, et al. Estrogen treatment improves arterial distensibility, fibrinolysis, and metabolic profile in postmenopausal women with type 2 diabetes mellitus. *Metabolism*. 2006;55(7). doi:10.1016/j.metabol.2006.03.003

48. Takato T, Yamada N, Ashida T. Effects of aging and sex on progression of carotid intima-media thickness: A retrospective 6-year follow-up study. *Geriatr Gerontol Int*. Published online 2008. doi:10.1111/j.1447-0594.2008.00467.x

49. Łoboz-Rudnicka M, Jaroch J, Bociąga Z, et al. Impact of cardiovascular risk factors on carotid intima-media thickness: Sex differences. *Clin Interv Aging*. Published online 2016. doi:10.2147/CIA.S103521

50. Laurent S, Cockcroft J, Van Bortel L, et al. Expert consensus document on arterial stiffness: Methodological issues and clinical applications. *Eur Heart J*. 2006;27(21). doi:10.1093/eurheartj/ehl254

51. O'Rourke MF, Staessen JA, Vlachopoulos C, Duprez D, Plante GE. Clinical applications of arterial stiffness; definitions and reference values. *Am J Hypertens*. 2002;15(5). doi:10.1016/S0895-7061(01)02319-6

52. Van Bortel LM, Kool MJ, Struijker Boudier HA. Effects of antihypertensive agents on local arterial distensibility and compliance. In: *Hypertension*. Vol 26. ; 1995. doi:10.1161/01.HYP.26.3.531

53. Godia EC, Madhok R, Pittman J, et al. Carotid artery distensibility: A reliability study. *J Ultrasound Med*. 2007;26(9). doi:10.7863/jum.2007.26.9.1157

54. Selzer RH, Mack WJ, Lee PL, Kwong-Fu H, Hodis HN. Improved common carotid elasticity and intima-media thickness measurements from computer analysis of sequential ultrasound frames. *Atherosclerosis*. 2001;154(1). doi:10.1016/S0021-9150(00)00461-5

55. Yuan C, Wang J, Ying M. Predictive value of carotid distensibility coefficient for cardiovascular diseases and all-cause mortality: A meta-analysis. *PLoS One*. 2016;11(4). doi:10.1371/journal.pone.0152799

56. Kazi AA, Blonde L. *Classification of Diabetes Mellitus. Geneva: World Health Organization; 2019*. Vol 21.; 2019.

57. Leow MKS. Glycated hemoglobin (HbA1c): Clinical applications of a mathematical concept. *Acta Inform Medica*. Published online 2016. doi:10.5455/aim.2016.24.233-238

58. WHO. *Definition and Diagnosis of Diabetes Mellitus and Intermediate Hyperglycemia: Report of a WHO/IDF Consultation*.; 2006.

59. World Health Organization. Report of a World Health Organization Consultation: Use of glycated haemoglobin (HbA1c) in the diagnosis of diabetes mellitus. *Diabetes Res Clin*

Pract. 2011;93(2).

60. Kahn SE, Cooper ME, Del Prato S. Pathophysiology and treatment of type 2 diabetes: Perspectives on the past, present, and future. *Lancet*. 2014;383(9922). doi:10.1016/S0140-6736(13)62154-6

61. Skyler JS, Bakris GL, Bonifacio E, et al. Differentiation of diabetes by pathophysiology, natural history, and prognosis. *Diabetes*. 2017;66(2). doi:10.2337/db16-0806

62. Furutani E. Closed-loop Blood Glucose Control for Type 1 Diabetes. *IEEJ Trans Electron Inf Syst*. 2019;139(4):260-263. doi:10.1541/ieejeiss.139.260

63. Ozougwu, J.C., Okimba, K.C., Belonwu, C.D., Unakalamba CB. The pathogenesis and pathophysiology of type 1 and type 2 diabetes mellitus. *J Physiol Pathophysiol*. Published online 2013. doi:10.5897/jpap2013.0001

64. Shepherd PR, Kahn BB. Glucose transporters and insulin action: Implications for insulin resistance and diabetes mellitus. *N Engl J Med*. Published online 1999. doi:10.1056/NEJM199907223410406

65. Zheng C, Liu Z. Vascular function, insulin action, and exercise: An intricate interplay. *Trends Endocrinol Metab*. Published online 2015. doi:10.1016/j.tem.2015.02.002

66. Alter P, Rupp H, Stoll F, et al. Increased enddiastolic wall stress precedes left ventricular hypertrophy in dilative heart failure - Use of the volume-based wall stress index. *Int J Cardiol*. 2012;157(2). doi:10.1016/j.ijcard.2011.07.092

67. Wang PJ, He F, Liao DH, et al. The impact of age on incremental elastic modulus and incremental compliance of pig hepatic portal vein for liver xenotransplantation. *Xenotransplantation*. 2009;16(1). doi:10.1111/j.1399-3089.2008.00505.x

68. Tynjälä A, Forsblom C, Harjutsalo V, Groop PH, Gordin D. Arterial stiffness predicts mortality in individuals with type 1 diabetes. *Diabetes Care*. 2020;43(9). doi:10.2337/dc20-0078

69. Zeitler P, Hirst K, Pyle L, et al. A Clinical Trial to Maintain Glycemic Control in Youth with Type 2 Diabetes The members of the writing group. *n engl j med*. 2012;24:2247-2256. doi:10.1056/NEJMoa1109333

70. Stumvoll M, Goldstein BJ, Van Haeften TW. Type 2 diabetes: Principles of pathogenesis and therapy. In: *Lancet*. Vol 365. ; 2005. doi:10.1016/S0140-6736(05)61032-X

71. Bogardus C, Lillioja S, Mott DM, Hollenbeck C, Reaven G. Relationship between degree of obesity and in vivo insulin action in man. *Am J Physiol - Endocrinol Metab*. 1985;11(3). doi:10.1152/ajpendo.1985.248.3.e286

72. Mooy JM, Grootenhuis PA, De Vries H, et al. Prevalence and determinants of glucose intolerance in a Dutch Caucasian population: The Hoorn study. *Diabetes Care*. 1995;18(9). doi:10.2337/diacare.18.9.1270

73. World Health Organization W. *Classification of Diabetes Mellitus. Geneva: World Health Organization; 2019. Licence: CC BY-NC-SA 3.0 IGO.*; 2019.

74. Cameron JD, Bulpitt CJ, Pinto ES, Rajkumar C. The aging of elastic and muscular arteries: A comparison of diabetic and nondiabetic subjects. *Diabetes Care*. 2003;26(7).

doi:10.2337/diacare.26.7.2133

75. Charvat J, Chlumsky J, Zakovicova E, Kvapil M. *Common Carotid Artery Intima-Media Thickness Is Not Increased but Distensibility Is Reduced in Normotensive Patients with Type 2 Diabetes Compared with Control Subjects*. Vol 38.; 2010.

76. Huang PL. A comprehensive definition for metabolic syndrome. *DMM Dis Model Mech*. 2009;2(5-6). doi:10.1242/dmm.001180

77. Park S, Lakatta EG. Role of inflammation in the pathogenesis of arterial stiffness. *Yonsei Med J*. 2012;53(2):258-261. doi:10.3349/ymj.2012.53.2.258

78. Nakhai-Pour HR, Grobbee DE, Bots ML, Muller M, Van der Schouw YT. C-reactive protein and aortic stiffness and wave reflection in middle-aged and elderly men from the community. *J Hum Hypertens*. 2007;21(12):949-955. doi:10.1038/sj.jhh.1002255

79. King DE, Mainous AG, Buchanan TA, Pearson WS. C-reactive protein and glycemic control in adults with diabetes. *Diabetes Care*. 2003;26(5):1535-1539. doi:10.2337/diacare.26.5.1535

80. Benigni A, Cassis P, Remuzzi G. Angiotensin II revisited: New roles in inflammation, immunology and aging. *EMBO Mol Med*. Published online 2010. doi:10.1002/emmm.201000080

81. Little RR, Sacks DB. HbA1c: How do we measure it and what does it mean? *Curr Opin Endocrinol Diabetes Obes*. 2009;16(2). doi:10.1097/MED.0b013e328327728d

82. Selvin E, Marinopoulos S, Berkenblit G, et al. Meta-analysis: Glycosylated hemoglobin and cardiovascular disease in diabetes mellitus. *Ann Intern Med*. 2004;141(6). doi:10.7326/0003-4819-141-6-200409210-00007

83. Zhang Y, Hu G, Yuan Z, Chen L. Glycosylated hemoglobin in relationship to cardiovascular outcomes and death in patients with type 2 diabetes: A systematic review and meta-analysis. *PLoS One*. 2012;7(8). doi:10.1371/journal.pone.0042551

84. Mannucci E, Dicembrini I, Lauria A, Pozzilli P. Is glucose control important for prevention of cardiovascular disease in diabetes? *Diabetes Care*. 2013;36(SUPPL.2). doi:10.2337/dcS13-2018

85. Moreno B, De Faria AP, Ritter AMV, et al. Glycated hemoglobin correlates with arterial stiffness and endothelial dysfunction in patients with resistant hypertension and uncontrolled diabetes mellitus. *J Clin Hypertens*. 2018;20(5). doi:10.1111/jch.13293

86. Prasad K. C-Reactive Protein and Cardiovascular Diseases. doi:10.1007/s00547-003-1018-y

87. Shrivastava AK, Singh HV, Raizada A, Singh SK. C-reactive protein, inflammation and coronary heart disease. *Egypt Hear J*. 2015;67(2). doi:10.1016/j.ehj.2014.11.005

88. McEniery CM, Wallace S, Mackenzie IS, Cockcroft JR, Wilkinson IB. C-Reactive Protein Is Associated With Arterial Stiffness in Apparently Healthy Individuals. Published online 2004. doi:10.1161/01.ATV.zhq0504.0173

89. Mattace-Raso FUS, Van Der Cammen TJM, Van Der Meer IM, et al. C-reactive protein and arterial stiffness in older adults: The Rotterdam Study. *Atherosclerosis*. 2004;176(1).

doi:10.1016/j.atherosclerosis.2004.04.014

90. Anderson TJ, Grégoire J, Pearson GJ, et al. 2016 Canadian Cardiovascular Society Guidelines for the Management of Dyslipidemia for the Prevention of Cardiovascular Disease in the Adult. *Can J Cardiol*. 2016;32(11). doi:10.1016/j.cjca.2016.07.510

91. Talayero BG, Sacks FM. The role of triglycerides in atherosclerosis. *Curr Cardiol Rep*. 2011;13(6). doi:10.1007/s11886-011-0220-3

92. Wang X, Ye P, Cao R, et al. Triglycerides are a predictive factor for arterial stiffness: a community-based 4.8-year prospective study. Published online 2016. doi:10.1186/s12944-016-0266-8

93. Alexopoulos AS, Qamar A, Hutchins K, Crowley MJ, Batch BC, Guyton JR. Triglycerides: Emerging Targets in Diabetes Care? Review of Moderate Hypertriglyceridemia in Diabetes. *Curr Diab Rep*. 2019;19(4). doi:10.1007/s11892-019-1136-3

94. Lemieux I, Lamarche B, Couillard C, et al. Total cholesterol/HDL cholesterol ratio vs LDL cholesterol/HDL cholesterol ratio as indices of ischemic heart disease risk in men. *Arch Intern Med*. 2001;161(22). doi:10.1001/archinte.161.22.2685

95. Wang F, Ye P, Luo L, et al. Association of serum lipids with arterial stiffness in a population-based study in Beijing. *Eur J Clin Invest*. 2011;41(9). doi:10.1111/j.1365-2362.2011.02481.x

96. Ference BA, Ginsberg HN, Graham I, et al. Low-density lipoproteins cause atherosclerotic cardiovascular disease. 1. Evidence from genetic, epidemiologic, and clinical studies. A consensus statement fromthe European Atherosclerosis Society Consensus Panel. *Eur Heart J*. 2017;38(32). doi:10.1093/eurheartj/ehx144

97. Barter P. The role of HDL-cholesterol in preventing atherosclerotic disease. In: *European Heart Journal, Supplement*. Vol 7. ; 2005. doi:10.1093/eurheartj/sui036

98. Jukema JW, Liem A-H, Dunselman PHJM, et al. LDL-C/HDL-C ratio in subjects with cardiovascular disease and a low HDL-C: results of the RADAR (Rosuvastatin and Atorvastatin in different Dosages And Reverse cholesterol transport) study LDL-C/HDL-C ratio in subjects with cardiovascular disease and a low HDL-C: results of the RADAR (Rosuvastatin and Atorvastatin in different Dosages Atorvastatin in different Dosages And Reverse cholesterol And Reverse cholesterol transport) study. 2005;21(11):1865-1874. doi:10.1185/030079905X74952

99. Prince CT, Secrest AM, Mackey RH, Arena VC, Kingsley LA, Orchard TJ. Cardiovascular autonomic neuropathy, HDL cholesterol, and smoking correlate with arterial stiffness markers determined 18 years later in type 1 diabetes. *Diabetes Care*. 2010;33(3). doi:10.2337/dc09-1936

100. Cao Y, Yan L, Guo N, et al. Non-high-density lipoprotein cholesterol and risk of cardiovascular disease in the general population and patients with type 2 diabetes: A systematic review and meta-analysis. *Diabetes Res Clin Pract*. 2019;147. doi:10.1016/j.diabres.2018.11.002

101. Colhoun HM, Betteridge DJ, Durrington PN, et al. Primary prevention of cardiovascular

disease with atorvastatin in type 2 diabetes in the Collaborative Atorvastatin Diabetes Study (CARDS): Multicentre randomised placebo-controlled trial. *Lancet*. 2004;364(9435). doi:10.1016/S0140-6736(04)16895-5

102. Flack JM, Adekola B. Blood pressure and the new ACC/AHA hypertension guidelines. *Trends Cardiovasc Med*. 2020;30(3). doi:10.1016/j.tcm.2019.05.003

103. Safar ME. Systolic blood pressure, pulse pressure and arterial stiffness as cardiovascular risk factors. *Curr Opin Nephrol Hypertens*. 2001;10(2). doi:10.1097/00041552-200103000-00015

104. DHHS. National Diabetes Statistics Report, 2020. *Natl Diabetes Stat Rep*. Published online 2020.

105. Forouzanfar M, Dajani HR, Groza VZ, Bolic M, Rajan S, Batkin I. Oscillometric blood pressure estimation: Past, present, and future. *IEEE Rev Biomed Eng*. 2015;8. doi:10.1109/RBME.2015.2434215

106. Rutan GH, Kuller LH, Neaton JD, Wentworth DN, McDonald RH, McFate Smith W. Mortality associated with diastolic hypertension and isolated systolic hypertension among men screened for the Multiple Risk Factor Intervention Trial. *Circulation*. 1988;77(3). doi:10.1161/01.CIR.77.3.504

107. Strandberg TE, Pitkala K. What is the most important component of blood pressure: Systolic, diastolic or pulse pressure? *Curr Opin Nephrol Hypertens*. 2003;12(3). doi:10.1097/00041552-200305000-00011

108. Pinto E. Blood pressure and ageing. *Postgrad Med J*. 2007;83(976). doi:10.1136/pgmj.2006.048371

109. Webb AJS. Progression of Arterial Stiffness is Associated With Midlife Diastolic Blood Pressure and Transition to Late-Life Hypertensive Phenotypes. *J Am Heart Assoc*. 2020;9(1). doi:10.1161/JAHA.119.014547

110. Grossman E. Does increased oxidative stress cause hypertension? *Diabetes Care*. 2008;31 Suppl 2. doi:10.2337/dc08-s246

111. Dharmashankar K, Widlansky ME. Vascular endothelial function and hypertension: Insights and directions. *Curr Hypertens Rep*. 2010;12(6). doi:10.1007/s11906-010-0150-2

112. Schulz E, Gori T, Münzel T. Oxidative stress and endothelial dysfunction in hypertension. *Hypertens Res*. 2011;34(6). doi:10.1038/hr.2011.39

113. Nuttall FQ. Body mass index: Obesity, BMI, and health: A critical review. *Nutr Today*. 2015;50(3). doi:10.1097/NT.0000000000000092

114. Khan SS, Ning H, Wilkins JT, et al. Association of Body Mass Index With Lifetime Risk of Cardiovascular Disease and Compression of Morbidity Supplemental content. *JAMA Cardiol*. 2018;3(4):280-287. doi:10.1001/jamacardio.2018.0022

115. Strasser B, Arvandi M, Pasha EP, Haley AP, Stanforth P, Tanaka H. Abdominal obesity is associated with arterial stiffness in middle-aged adults. *Nutr Metab Cardiovasc Dis*. 2015;25(5). doi:10.1016/j.numecd.2015.01.002

116. Carbone S, Canada JM, Billingsley HE, Siddiqui MS, Elagizi A, Lavie CJ. Obesity

paradox in cardiovascular disease: Where do we stand? *Vasc Health Risk Manag*. 2019;15. doi:10.2147/VHRM.S168946

117. Tanaka H, DeSouza CA, Seals DR. Absence of age-related increase in central arterial stiffness in physically active women. *Arterioscler Thromb Vasc Biol*. Published online 1998. doi:10.1161/01.ATV.18.1.127

118. Schmitz KH, Arnett DK, Bank A, et al. Arterial distensibility and physical activity in the ARIC study. *Med Sci Sports Exerc*. 2001;33(12). doi:10.1097/00005768-200112000-00014

119. Minder CM, Shaya GE, Michos ED, et al. Relation between self-reported physical activity level, fitness, and cardiometabolic risk. *Am J Cardiol*. 2014;113(4). doi:10.1016/j.amjcard.2013.11.010

120. Moreau KL, Silver AE, Dinenno FA, Seals DR. Habitual aerobic exercise is associated with smaller femoral artery intima media thickness with age in healthy men and women. *Eur J Prev Cardiol*. Published online 2006. doi:10.1097/01.hjr.0000230103.55653.42

121. Gando Y, Yamamoto K, Kawano H, et al. Attenuated age-related carotid arterial remodeling in adults with a high level of cardiorespiratory fitness. *J Atheroscler Thromb*. Published online 2011. doi:10.5551/jat.6924

122. Kozakova M, Palombo C, Mhamdi L, et al. Habitual physical activity and vascular aging in a young to middle-age population at low cardiovascular risk. *Stroke*. Published online 2007. doi:10.1161/STROKEAHA.107.484949

123. Baecke JAH, Burema J, Frijters JER. A short questionnaire for the measurement of habitual physical activity in epidemiological studies. *Am J Clin Nutr*. 1982;36(5). doi:10.1093/ajcn/36.5.936

124. Cotie LM, Josse AR, Philips SM, MacDonald MJ. 16-Weeks of Combined Aerobic and Resistance Training and Hypo-Caloric Diet on Measures of Arterial Stiffness in Overweight Pre-Menopausal Women. *J Metab Syndr*. 2014;03(01). doi:10.4172/2167-0943.1000137

125. Cotie LM, Currie KD, McGill GM, et al. Associations between measures of vascular structure and function and systemic circulating blood markers in humans. *Physiol Rep*. 2016;4(18). doi:10.14814/phy2.12982

126. Königstein K, Infanger D, Klenk C, Carrard J, Hinrichs T, Schmidt-Trucksäss A. Physical activity is favorably associated with arterial stiffness in patients with obesity and elevated metabolic risk. *Int J Clin Pract*. 2020;74(9). doi:10.1111/ijcp.13563

127. Stamatelopoulos K, Tsoltos N, Armeni E, et al. Physical activity is associated with lower arterial stiffness in normal-weight postmenopausal women. *J Clin Hypertens*. 2020;22(9). doi:10.1111/jch.13954

128. Raina PS, Wolfson C, Kirkland SA, et al. The canadian longitudinal study on aging (CLSA). *Can J Aging*. 2009;28(3):221-229. doi:10.1017/S0714980809990055

129. Raina P, Wolfson C, Kirkland S, et al. Cohort Profile: The Canadian Longitudinal Study on Aging (CLSA). *Int J Epidemiol*. 2019;48(6):1752-1753J. doi:10.1093/ije/dyz173

130. Raina P, Wolfson C, Kirkland S, Griffith L, Griffi L. The Canadian Longitudinal Study on Aging (CLSA) Report on Health and Aging in Canada - Findings from Baseline Data Collection. Published online 2018. https://www.clsa-elcv.ca/doc/2639

131. Maintaining Contact Questionnaire (Tracking and Comprehensive): CLSA Scientific Working Groups. Published online 2015. https://clsa-elcv.ca/doc/540

132. Washburn RA, Smith KW, Jette AM, Janney CA. The physical activity scale for the elderly (PASE): Development and evaluation. *J Clin Epidemiol*. 1993;46(2). doi:10.1016/0895-4356(93)90053-4

133. Choi HL, Au JS, MacDonald MJ. Carotid extra-media thickness increases with age, but is not related to arterial stiffness in adults. *Artery Res*. 2018;21. doi:10.1016/j.artres.2017.12.003

134. Kamruzzaman MD, M Rahmatullah Imon AH. High leverage point: another source of multicollinearity. *Pakistan J Stat.* 2002;(January 2002).

135. Cook RD, Weisberg S. Residuals and Influence in Regression. In: ; 1982.

136. Iacobucci D, Schneider MJ, Popovich DL, Bakamitsos GA. Mean centering helps alleviate "micro" but not "macro" multicollinearity. *Behav Res Methods*. 2016;48(4). doi:10.3758/s13428-015-0624-x

137. Iacobucci D, Schneider MJ, Popovich DL, Bakamitsos GA. Mean centering, multicollinearity, and moderators in multiple regression: The reconciliation redux. *Behav Res Methods*. 2017;49(1). doi:10.3758/s13428-016-0827-9

138. Tsuchikura S, Shoji T, Kimoto E, et al. Central versus peripheral arterial stiffness in association with coronary, cerebral and peripheral arterial disease. *Atherosclerosis*. 2010;211(2). doi:10.1016/j.atherosclerosis.2010.03.037

139. Logan SL, Gottlieb BH, Maitl SB, Meegan D, Spriet LL. The physical activity scale for the elderly (PASE) questionnaire; Does it predict physical health? *Int J Environ Res Public Health*. Published online 2013. doi:10.3390/ijerph10093967

140. Smulyan H, Lieber A, Safar ME. Hypertension, Diabetes Type II, and their association: Role of arterial stiffness. *Am J Hypertens*. 2016;29(1). doi:10.1093/ajh/hpv107

141. Fox CS, Sullivan L, D'Agostino RB, Wilson PWF. The Significant Effect of Diabetes Duration on Coronary Heart Disease Mortality: The Framingham Heart Study. *Diabetes Care*. 2004;27(3). doi:10.2337/diacare.27.3.704

142. Spijkerman AMW, Dekker JM, Nijpels G, et al. Impact of diabetes duration and cardiovascular risk factors on mortality in type 2 diabetes: The Hoorn Study. *Eur J Clin Invest*. 2002;32(12). doi:10.1046/j.1365-2362.2002.01090.x

143. Vaidya D, Heckbert SR, Wasserman BA, Ouyang P. Sex-specific association of age with carotid artery distensibility: Multi-ethnic study of atherosclerosis. *J Women's Heal*. 2012;21(5). doi:10.1089/jwh.2011.3220

144. De Angelis L, Millasseau SC, Smith A, et al. Sex differences in age-related stiffening of the aorta in subjects with type 2 diabetes. *Hypertension*. 2004;44(1). doi:10.1161/01.HYP.0000130482.81883.fd

145. Mitchell GF. Arterial stiffness and hypertension: Chicken or egg? *Hypertension*. 2014;64(2). doi:10.1161/HYPERTENSIONAHA.114.03449

146. McGorrian C, Yusuf S, Islam S, et al. Estimating modifiable coronary heart disease risk in multiple regions of the world: The INTERHEART Modifiable Risk Score. *Eur Heart J*. 2011;32(5). doi:10.1093/eurheartj/ehq448

147. Yusuf S, Rangarajan S, Teo K, et al. Cardiovascular Risk and Events in 17 Low-, Middle-, and High-Income Countries. *N Engl J Med*. 2014;371(9). doi:10.1056/nejmoa1311890

148. Privšek E, Hellgren M, Råstam L, Lindblad U, Daka B. Epidemiological and clinical implications of blood pressure measured in seated versus supine position. *Med (United States)*. 2018;97(31). doi:10.1097/MD.0000000000011603

APPENDIX A: SPSS OUTPUT

Descriptive Statistics

Statistics

		Age	Age of diagnosis	Distensibility	cIMT	BMI	hs-CRP	HBA1c
N	Valid	2201	2189	2201	2015	2196	2201	2201
	Missing	0	12	0	186	5	0	0
Mean		65.08	55.5007	.00215116076 1	.7599799 8	29.4698	2.762	6.423
Median		65.00	57.0000	.00199782300 0	.7398850 0	28.9331	1.300	6.100
Std. Deviation		9.629	14.16257	.00102590186 96	.1715752 75	5.23280	5.3238	1.1973
Skewness		.031	-.745	1.319	.715	.799	8.822	2.057
Std. Error of Skewness		.052	.052	.052	.055	.052	.052	.052
Kurtosis		-.817	.752	3.601	1.263	1.316	112.465	6.193
Std. Error of Kurtosis		.104	.105	.104	.109	.104	.104	.104
Minimum		45	.00	.0003371460	.235172	15.76	.2	4.7
Maximum		85	85.00	.0088971900	1.654660	54.88	101.1	14.2
Percentiles	25	58.00	49.0000	.00142479550 0	.6390370 0	25.7357	.700	5.600
	50	65.00	57.0000	.00199782300 0	.7398850 0	28.9331	1.300	6.100
	75	72.00	65.0000	.00270264300 0	.8585720 0	32.3608	2.900	6.800

Statistics

		Triglycerides	HDL-C	LDL-C	Cholesterol	TC/HDL Ratio	Systolic Blood Pressure
N	Valid	2201	2201	2121	2201	2201	2201
	Missing	0	0	80	0	0	0
Mean		1.9872	1.3522	2.4105	4.6492	3.7099	123.362032409510770
Median		1.7000	1.2800	2.2500	4.5000	3.4366	121.600000000000000
Std. Deviation		1.11438	.44248	.99849	1.17890	1.33115	16.178066660996180
Skewness		1.641	1.038	.611	.536	1.272	.659
Std. Error of Skewness		.052	.052	.053	.052	.052	.052
Kurtosis		3.946	1.669	.090	.006	2.245	1.250
Std. Error of Kurtosis		.104	.104	.106	.104	.104	.104
Minimum		.39	.41	.14	1.80	1.43	70.8000000000000
Maximum		9.44	3.70	6.54	9.02	12.07	199.0000000000000
Percentiles	25	1.2200	1.0300	1.6600	3.7500	2.7564	112.600000000000000
	50	1.7000	1.2800	2.2500	4.5000	3.4366	121.600000000000000
	75	2.4550	1.5900	3.0900	5.4200	4.2972	132.700000000000000

Statistics

		Diastolic Blood Pressure	Maximal Grip Strength	PASE score	Get up and Go Test	4-metre walk Time	Chair rise test Time
N	Valid	2201	2052	2073	2184	2190	2091
	Missing	0	149	128	17	11	110
Mean		73.419218537028640	34.9292	131.3622	10.0396	4.4215	13.8720
Median		73.400000000000000	33.8638	119.5714	9.6500	4.2200	13.4000
Std. Deviation		10.044522474591481	11.17756	69.33213	2.39253	1.04370	3.73437
Skewness		.243	.395	.825	2.166	2.083	1.387
Std. Error of Skewness		.052	.054	.054	.052	.052	.054
Kurtosis		.250	-.380	.612	10.328	10.919	6.115
Std. Error of Kurtosis		.104	.108	.107	.105	.105	.107
Minimum		44.800000000000	9.21	2.14	3.22	1.97	3.66
Maximum		119.000000000000	71.87	427.18	30.16	15.13	44.10
Percentiles	25	66.300000000000010	25.9318	82.1429	8.5350	3.7200	11.3700
	50	73.400000000000000	33.8638	119.5714	9.6500	4.2200	13.4000
	75	79.600000000000000	42.8182	171.0000	11.0300	4.9100	15.8200

Independent Samples T-test for Normal Data; Stratified by Sex

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means		
		F	Sig.	t	df	Significance One-Sided p
Age	Equal variances assumed	2.443	.118	-2.774	2199	.003
	Equal variances not assumed			-2.760	2005.191	.003
cIMT	Equal variances assumed	8.347	.004	-3.680	2013	<.001
	Equal variances not assumed			-3.723	1885.115	<.001
BMI	Equal variances assumed	79.478	<.001	3.580	2194	<.001
	Equal variances not assumed			3.453	1714.820	<.001
LDL-C	Equal variances assumed	6.993	.008	9.252	2119	<.001
	Equal variances not assumed			9.162	1913.690	<.001
Cholesterol	Equal variances assumed	6.977	.008	13.916	2199	<.001
	Equal variances not assumed			13.773	1958.628	<.001
Systolic Blood Pressure	Equal variances assumed	1.254	.263	-2.599	2199	.005
	Equal variances not assumed			-2.590	2015.019	.005
Diastolic Blood Pressure	Equal variances assumed	.563	.453	-8.272	2199	<.001
	Equal variances not assumed			-8.325	2088.873	<.001
Maximal Grip Strength	Equal variances assumed	134.813	<.001	-41.689	2050	<.001
	Equal variances not assumed			-44.436	2023.272	<.001
4-metre Walk Time	Equal variances assumed	3.513	.061	2.664	2188	.004

	Equal variances not assumed			2.637	1953.268	.004
Chair Rise Test Time	Equal variances assumed	4.092	.043	1.539	2089	.062
	Equal variances not assumed			1.521	1843.685	.064

Independent Samples Test

		t-test for Equality of Means			
		Significance Two-Sided p	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference Lower
Age	Equal variances assumed	.006	-1.148	.414	-1.959
	Equal variances not assumed	.006	-1.148	.416	-1.963
cIMT	Equal variances assumed	<.001	-.028424550	.007724434	-.043573271
	Equal variances not assumed	<.001	-.028424550	.007635560	-.043399589
BMI	Equal variances assumed	<.001	.80493	.22484	.36401
	Equal variances not assumed	<.001	.80493	.23311	.34772
LDL-C	Equal variances assumed	<.001	.39645	.04285	.31242
	Equal variances not assumed	<.001	.39645	.04327	.31159

Cholesterol	Equal variances assumed	<.001	.67700	.04865	.58160
	Equal variances not assumed	<.001	.67700	.04915	.58060
Systolic Blood Pressure	Equal variances assumed	.009	-1.807446968740976	.6953102973 96581	-3.170980613328140
	Equal variances not assumed	.010	-1.807446968740976	.6978138836 16988	-3.175959066368648
Diastolic Blood Pressure	Equal variances assumed	<.001	-3.522040052168648	.4257885187 90797	-4.357029801384235
	Equal variances not assumed	<.001	-3.522040052168648	.4230727569 27193	-4.351728163574751
Maximal Grip Strength	Equal variances assumed	<.001	-15.32623	.36763	-16.04720
	Equal variances not assumed	<.001	-15.32623	.34491	-16.00264
4-metre Walk Time	Equal variances assumed	.008	.11975	.04496	.03159
	Equal variances not assumed	.008	.11975	.04541	.03068
Chair Rise Test Time	Equal variances assumed	.124	.25383	.16493	-.06961
	Equal variances not assumed	.128	.25383	.16683	-.07337

Independent Samples Test

		t-test for Equality of Means 95% Confidence Interval of the Difference Upper
Age	Equal variances assumed	-.336
	Equal variances not assumed	-.332
cIMT	Equal variances assumed	-.013275830
	Equal variances not assumed	-.013449512
BMI	Equal variances assumed	1.24585
	Equal variances not assumed	1.26215
LDL-C	Equal variances assumed	.48049
	Equal variances not assumed	.48132
Cholesterol	Equal variances assumed	.77240
	Equal variances not assumed	.77340
Systolic Blood Pressure	Equal variances assumed	-.443913324153811
	Equal variances not assumed	-.438934871113304
Diastolic Blood Pressure	Equal variances assumed	-2.687050302953062
	Equal variances not assumed	-2.692351940762545
Maximal Grip Strength	Equal variances assumed	-14.60526
	Equal variances not assumed	-14.64982
4-metre Walk Time	Equal variances assumed	.20791
	Equal variances not assumed	.20882
Chair Rise Test Time	Equal variances assumed	.57727

	Equal variances not assumed	.58103

Mann-Whitney U Non-Parametric Independent Samples Test for Non-Normal Data; Stratified by Sex

Hypothesis Test Summary

	Null Hypothesis	Test	Sig.[a,b]	Decision
1	The distribution of Age of diagnosis_ is the same across categories of Sex.	Independent-Samples Mann-Whitney U Test	.002	Reject the null hypothesis.
2	The distribution of Distensibility is the same across categories of Sex.	Independent-Samples Mann-Whitney U Test	.972	Retain the null hypothesis.
3	The distribution of hs-CRP is the same across categories of Sex.	Independent-Samples Mann-Whitney U Test	.000	Reject the null hypothesis.
4	The distribution of HBA1c is the same across categories of Sex.	Independent-Samples Mann-Whitney U Test	<.001	Reject the null hypothesis.
5	The distribution of Triglycerides is the same across categories of Sex.	Independent-Samples Mann-Whitney U Test	.985	Retain the null hypothesis.
6	The distribution of HDL-C is the same across categories of Sex.	Independent-Samples Mann-Whitney U Test	.000	Reject the null hypothesis.
7	The distribution of TC/HDL Ratio is the same across categories of Sex/Gender.	Independent-Samples Mann-Whitney U Test	<.001	Reject the null hypothesis.
8	The distribution of PASE score is the same across categories of Sex/Gender.	Independent-Samples Mann-Whitney U Test	<.001	Reject the null hypothesis.
9	The distribution of Get up and Go Test is the same across categories of Sex.	Independent-Samples Mann-Whitney U Test	.707	Retain the null hypothesis.

a. The significance level is .050.
b. Asymptotic significance is displayed.

Age of Diagnosis across Sex/Gender

Independent-Samples Mann-Whitney U Test Summary

Total N	2189
Mann-Whitney U	632292.500
Wilcoxon W	1409173.500
Test Statistic	632292.500
Standard Error	14638.195
Standardized Test Statistic	3.061
Asymptotic Sig.(2-sided test)	.002

Distensibility across Sex/Gender

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	594743.000
Wilcoxon W	1377869.000
Test Statistic	594743.000
Standard Error	14767.551
Standardized Test Statistic	.035
Asymptotic Sig.(2-sided test)	.972

hs-CRP across Sex/Gender

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	466183.500
Wilcoxon W	1249309.500
Test Statistic	466183.500
Standard Error	14757.679
Standardized Test Statistic	-8.676
Asymptotic Sig.(2-sided test)	.000

HBA1c across Sex/Gender

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	689227.500
Wilcoxon W	1472353.500
Test Statistic	689227.500
Standard Error	14754.386
Standardized Test Statistic	6.439
Asymptotic Sig.(2-sided test)	<.001

Triglycerides across Sex/Gender

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	594510.000
Wilcoxon W	1377636.000
Test Statistic	594510.000
Standard Error	14767.414
Standardized Test Statistic	.019
Asymptotic Sig.(2-sided test)	.985

HDL-C across Sex/Gender

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	360159.500
Wilcoxon W	1143285.500
Test Statistic	360159.500
Standard Error	14767.037
Standardized Test Statistic	-15.851
Asymptotic Sig.(2-sided test)	.000

TC/HDL Ratio across Sex/Gender

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	660390.500
Wilcoxon W	1443516.500
Test Statistic	660390.500
Standard Error	14767.551
Standardized Test Statistic	4.480
Asymptotic Sig.(2-sided test)	<.001

PASE score across Sex/Gender

Independent-Samples Mann-Whitney U Test Summary

Total N	2073
Mann-Whitney U	606013.500
Wilcoxon W	1305166.500
Test Statistic	606013.500
Standard Error	13491.445
Standardized Test Statistic	5.888
Asymptotic Sig.(2-sided test)	<.001

Get Up and Go Test_ across Sex/Gender

Independent-Samples Mann-Whitney U Test Summary

Total N	2184
Mann-Whitney U	591055.000
Wilcoxon W	1357996.000
Test Statistic	591055.000
Standard Error	14602.851
Standardized Test Statistic	.375
Asymptotic Sig.(2-sided test)	.707

Independent Samples T-test for Normal Data; Stratified by Diabetes Type

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means		
		F	Sig.	t	df	Significance One-Sided p
Age	Equal variances assumed	.733	.392	2.405	2199	.008
	Equal variances not assumed			2.276	78.709	.013
cIMT	Equal variances assumed	.050	.822	.312	2013	.377
	Equal variances not assumed			.329	77.014	.372
BMI	Equal variances assumed	.260	.610	3.872	2194	<.001
	Equal variances not assumed			4.087	78.826	<.001
LDL-C	Equal variances assumed	18.319	<.001	3.503	2119	<.001
	Equal variances not assumed			4.744	82.200	<.001
Cholesterol	Equal variances assumed	15.512	<.001	2.818	2199	.002
	Equal variances not assumed			3.955	85.236	<.001
Systolic Blood Pressure	Equal variances assumed	.736	.391	.804	2199	.211
	Equal variances not assumed			.782	78.994	.218
Diastolic Blood Pressure	Equal variances assumed	.003	.956	3.302	2199	<.001
	Equal variances not assumed			3.314	79.353	<.001
Maximal Grip Strength	Equal variances assumed	6.419	.011	2.471	2050	.007
	Equal variances not assumed			2.921	71.317	.002
4-metre Walk Time	Equal variances assumed	1.718	.190	-.421	2188	.337
	Equal variances not assumed			-.462	80.556	.323

Chair Rise Test Time	Equal variances assumed	.489	.484	.424	2089	.336
	Equal variances not assumed			.477	75.277	.317

Independent Samples Test

		t-test for Equality of Means			
		Significance Two-Sided p	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference Lower
Age	Equal variances assumed	.016	2.718	1.130	.502
	Equal variances not assumed	.026	2.718	1.194	.341
cIMT	Equal variances assumed	.755	.006428303	.020596191	-.033963777
	Equal variances not assumed	.743	.006428303	.019535076	-.032470889
BMI	Equal variances assumed	<.001	2.38816	.61685	1.17848
	Equal variances not assumed	<.001	2.38816	.58436	1.22499
LDL-C	Equal variances assumed	<.001	.41546	.11861	.18285
	Equal variances not assumed	<.001	.41546	.08758	.24124
Cholesterol	Equal variances assumed	.005	.38970	.13829	.11851
	Equal variances not assumed	<.001	.38970	.09852	.19381
Systolic Blood Pressure	Equal variances assumed	.421	1.528793352148185	1.900899881143613	-2.198953745278599
	Equal variances not assumed	.437	1.528793352148185	1.955104751394411	-2.362749874470220
Diastolic Blood Pressure	Equal variances assumed	<.001	3.887693320790050	1.177475695653094	1.578612421631537
	Equal variances not assumed	.001	3.887693320790050	1.173132080271146	1.552793690880154

Maximal Grip Strength	Equal variances assumed	.014	3.45214	1.39680	.71285
	Equal variances not assumed	.005	3.45214	1.18200	1.09548
4-metre Walk Time	Equal variances assumed	.674	-.05162	.12266	-.29215
	Equal variances not assumed	.645	-.05162	.11165	-.27379
Chair Rise Test Time	Equal variances assumed	.671	.19275	.45410	-.69778
	Equal variances not assumed	.635	.19275	.40388	-.61177

Independent Samples Test

		t-test for Equality of Means
		95% Confidence Interval of the Difference
		Upper
Age	Equal variances assumed	4.934
	Equal variances not assumed	5.095
cIMT	Equal variances assumed	.046820382
	Equal variances not assumed	.045327494
BMI	Equal variances assumed	3.59784
	Equal variances not assumed	3.55133
LDL-C	Equal variances assumed	.64807
	Equal variances not assumed	.58968
Cholesterol	Equal variances assumed	.66089
	Equal variances not assumed	.58558

Systolic Blood Pressure	Equal variances assumed	5.256540449574969
	Equal variances not assumed	5.420336578766589
Diastolic Blood Pressure	Equal variances assumed	6.196774219948561
	Equal variances not assumed	6.222592950699944
Maximal Grip Strength	Equal variances assumed	6.19144
	Equal variances not assumed	5.80881
4-metre Walk Time	Equal variances assumed	.18892
	Equal variances not assumed	.17056
Chair Rise Test Time	Equal variances assumed	1.08327
	Equal variances not assumed	.99727

Mann-Whitney U Non-Parametric Independent Samples Test for Non-Normal Data; Stratified by Sex

Hypothesis Test Summary

	Null Hypothesis	Test	Sig.[a,b]	Decision
1	The distribution of Age of diagnosis is the same across categories of Type1 vs other.	Independent-Samples Mann-Whitney U Test	.000	Reject the null hypothesis.
2	The distribution of Distensibility is the same across categories of Type1 vs other.	Independent-Samples Mann-Whitney U Test	.792	Retain the null hypothesis.
3	The distribution of hs-CRP is the same across categories of Type1 vs other.	Independent-Samples Mann-Whitney U Test	.476	Retain the null hypothesis.
4	The distribution of HBA1c is the same across categories of Type1 vs other.	Independent-Samples Mann-Whitney U Test	.000	Reject the null hypothesis.

5	The distribution of Triglycerides is the same across categories of Type1 vs other.	Independent-Samples Mann-Whitney U Test	<.001	Reject the null hypothesis.
6	The distribution of HDL-C is the same across categories of Type1 vs other.	Independent-Samples Mann-Whitney U Test	<.001	Reject the null hypothesis.
7	The distribution of TC/HDL Ratio is the same across categories of Type1 vs other.	Independent-Samples Mann-Whitney U Test	<.001	Reject the null hypothesis.
8	The distribution of PASE score is the same across categories of Type1 vs other.	Independent-Samples Mann-Whitney U Test	.960	Retain the null hypothesis.
9	The distribution of Get up and Go Test is the same across categories of Type1 vs other.	Independent-Samples Mann-Whitney U Test	.562	Retain the null hypothesis.

a. The significance level is .050.
b. Asymptotic significance is displayed.

Independent-Samples Mann-Whitney U Test

Age of diagnosis across Type1 vs other

Independent-Samples Mann-Whitney U Test Summary

Total N	2189
Mann-Whitney U	31447.000
Wilcoxon W	34297.000
Test Statistic	31447.000
Standard Error	5377.195
Standardized Test Statistic	-8.895
Asymptotic Sig.(2-sided test)	.000

Distensibility across Type1 vs other

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	81154.000
Wilcoxon W	84004.000
Test Statistic	81154.000
Standard Error	5409.166
Standardized Test Statistic	.264
Asymptotic Sig.(2-sided test)	.792

hs-CRP across Type1 vs other

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	75874.000
Wilcoxon W	78724.000
Test Statistic	75874.000
Standard Error	5405.550
Standardized Test Statistic	-.712
Asymptotic Sig.(2-sided test)	.476

HBA1c across Type1 vs other

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	131319.500
Wilcoxon W	134169.500
Test Statistic	131319.500
Standard Error	5404.343
Standardized Test Statistic	9.547
Asymptotic Sig.(2-sided test)	.000

Triglycerides across Type1 vs other

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	49731.000
Wilcoxon W	52581.000
Test Statistic	49731.000
Standard Error	5409.116
Standardized Test Statistic	-5.545
Asymptotic Sig.(2-sided test)	<.001

HDL-C across Type1_vs_other

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	103431.000
Wilcoxon W	106281.000
Test Statistic	103431.000
Standard Error	5408.977
Standardized Test Statistic	4.383
Asymptotic Sig.(2-sided test)	<.001

TC/HDL Ratio across Type1 vs other

Independent-Samples Mann-Whitney U Test Summary

Total N	2201
Mann-Whitney U	45614.500
Wilcoxon W	48464.500
Test Statistic	45614.500
Standard Error	5409.166
Standardized Test Statistic	-6.306
Asymptotic Sig.(2-sided test)	<.001

PASE score across Type1 vs other

Independent-Samples Mann-Whitney U Test Summary

Total N	2073
Mann-Whitney U	69859.000
Wilcoxon W	72344.000
Test Statistic	69859.000
Standard Error	4922.666
Standardized Test Statistic	-.050
Asymptotic Sig.(2-sided test)	.960

Get up and Go Test across Type1 vs other

Independent-Samples Mann-Whitney U Test Summary

Total N	2184
Mann-Whitney U	81162.000
Wilcoxon W	83937.000
Test Statistic	81162.000
Standard Error	5331.984
Standardized Test Statistic	.580
Asymptotic Sig.(2-sided test)	.562

General Linear Model: Unadjusted Model 1 with Interactions (PASE, Age, and Sex)

Between-Subjects Factors

		N
Sex/Gender	0	857
	1	1146

Levene's Test of Equality of Error Variances[a]

Dependent Variable: Distensibility

F	df1	df2	Sig.
.769	1	2001	.381

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.[a]

a. Design: Intercept + PASE_centred + Sex_A + Age + Sex_A * PASE_centred + PASE_centred * Age + Sex_A * Age

Tests for Heteroskedasticity

White Test for Heteroskedasticity[a,b,c]

Chi-Square	df	Sig.
10.566	16	.835

a. Dependent variable: Distensibility

b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.

c. Design: Intercept + PASE_centred + Sex_A + Age + Sex_A * PASE_centred + PASE_centred * Age + Sex_A * Age + PASE_centred * PASE_centred + Sex_A * PASE_centred * PASE_centred + PASE_centred * PASE_centred * Age + Sex_A * PASE_centred * Age + Age * Age + PASE_centred * Age * Age + Sex_A * Age * Age + Sex_A * PASE_centred * PASE_centred * Age + PASE_centred * PASE_centred * Age * Age + Sex_A * PASE_centred * Age * Age

Modified Breusch-Pagan Test for Heteroskedasticity[a,b,c]

Chi-Square	df	Sig.
2.699	1	.100

a. Dependent variable: Distensibility

b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.

c. Predicted values from design: Intercept + PASE_centred + Sex_A + Age + Sex_A * PASE_centred + PASE_centred * Age + Sex_A * Age

Breusch-Pagan Test for Heteroskedasticity[a,b,c]

Chi-Square	df	Sig.
8.975	1	.003

a. Dependent variable: Distensibility
b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.
c. Predicted values from design: Intercept + PASE_centred + Sex_A + Age + Sex_A * PASE_centred + PASE_centred * Age + Sex_A * Age

F Test for Heteroskedasticity[a,b,c]

F	df1	df2	Sig.
2.700	1	2001	.101

a. Dependent variable: Distensibility
b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.
c. Predicted values from design: Intercept + PASE_centred + Sex_A + Age + Sex_A * PASE_centred + PASE_centred * Age + Sex_A * Age

Tests of Between-Subjects Effects

Dependent Variable: Distensibility

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	.000[a]	6	3.013E-5	31.405	<.001	.086
Intercept	.001	1	.001	661.560	<.001	.249
PASE_centred	6.832E-7	1	6.832E-7	.712	.399	.000
Sex_A	3.107E-6	1	3.107E-6	3.238	.072	.002
Age	.000	1	.000	157.518	<.001	.073
Sex_A * PASE_centred	2.231E-8	1	2.231E-8	.023	.879	.000
PASE_centred * Age	6.241E-7	1	6.241E-7	.650	.420	.000
Sex_A * Age	3.479E-6	1	3.479E-6	3.626	.057	.002
Error	.002	1996	9.595E-7			
Total	.011	2003				
Corrected Total	.002	2002				

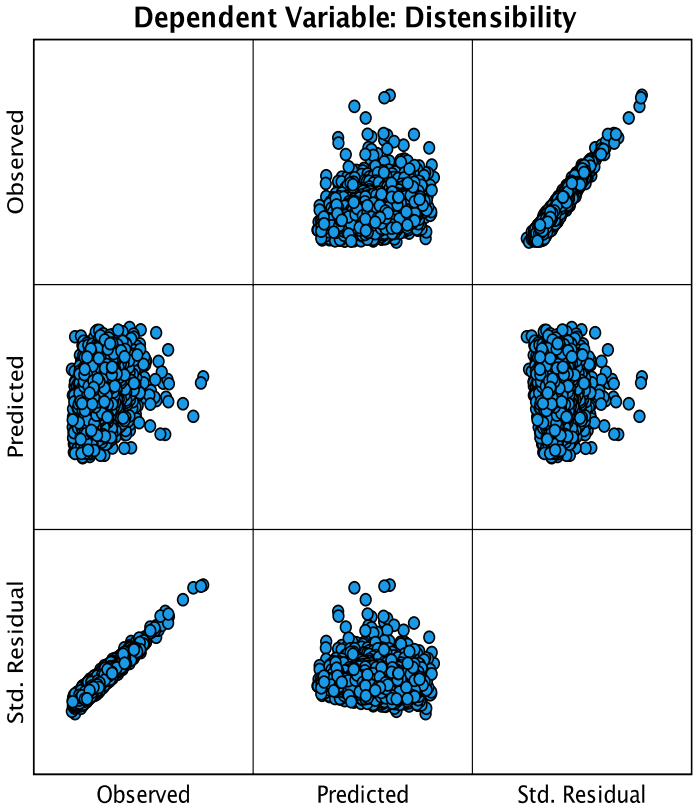
a. R Squared = .086 (Adjusted R Squared = .084)

Parameter Estimates

Dependent Variable: Distensibility

Parameter	B	Std. Error	t	Sig.	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound	Partial Eta Squared
Intercept	.004	.000	17.959	<.001	.003	.004	.139
PASE_centred	1.882E-6	2.220E-6	.848	.397	-2.472E-6	6.237E-6	.000
[Sex_A=0]	.001	.000	1.799	.072	-5.292E-5	.001	.002
[Sex_A=1]	0[a]	.	.	.	.	.	.
Age	-2.683E-5	3.274E-6	-8.196	<.001	-3.325E-5	-2.041E-5	.033
[Sex_A=0] * PASE_centred	-1.088E-7	7.133E-7	-.152	.879	-1.508E-6	1.290E-6	.000
[Sex_A=1] * PASE_centred	0[a]	.	.	.	.	.	.
PASE_centred * Age	-2.815E-8	3.491E-8	-.807	.420	-9.661E-8	4.030E-8	.000
[Sex_A=0] * Age	-9.532E-6	5.006E-6	-1.904	.057	-1.935E-5	2.855E-7	.002
[Sex_A=1] * Age	0[a]	.	.	.	.	.	.

a. This parameter is set to zero because it is redundant.



Model: Intercept + PASE_centred + Sex_A + Age + Sex_A PASE_centred + PASE_centred * Age + Sex_A Age

General Linear Model: Unadjusted Model 2 with No Interactions and Outliers Removed (PASE, Age, and Sex)

Univariate Analysis of Variance

Between-Subjects Factors

		N
Sex/Gender	0	846
	1	1133

Levene's Test of Equality of Error Variances[a]

Dependent Variable: Distensibility

F	df1	df2	Sig.
1.433	1	1977	.231

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.[a]

a. Design: Intercept + PASE_centred + Sex_A + Age

Tests for Heteroskedasticity

White Test for Heteroskedasticity[a,b,c]

Chi-Square	df	Sig.
26.281	8	<.001

a. Dependent variable: Distensibility
b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.
c. Design: Intercept + PASE_centred + Sex_A + Age + PASE_centred * PASE_centred + Sex_A * PASE_centred + PASE_centred * Age + Sex_A * Age + Age * Age

Modified Breusch-Pagan Test for Heteroskedasticity[a,b,c]

Chi-Square	df	Sig.
15.071	1	<.001

a. Dependent variable: Distensibility
b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.
c. Predicted values from design: Intercept + PASE_centred + Sex_A + Age

Breusch-Pagan Test for Heteroskedasticity[a,b,c]

Chi-Square	df	Sig.
16.706	1	<.001

a. Dependent variable: Distensibility
b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.
c. Predicted values from design: Intercept + PASE_centred + Sex_A + Age

F Test for Heteroskedasticity[a,b,c]

F	df1	df2	Sig.
15.171	1	1977	<.001

a. Dependent variable: Distensibility
b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.
c. Predicted values from design: Intercept + PASE_centred + Sex_A + Age

Tests of Between-Subjects Effects

Dependent Variable: Distensibility

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	.000[a]	3	5.874E-5	79.024	<.001	.107
Intercept	.001	1	.001	805.329	<.001	.290
PASE_centred	1.597E-6	1	1.597E-6	2.148	.143	.001
Sex_A	3.380E-7	1	3.380E-7	.455	.500	.000
Age	.000	1	.000	188.852	<.001	.087
Error	.001	1975	7.433E-7			
Total	.010	1979				
Corrected Total	.002	1978				

a. R Squared = .107 (Adjusted R Squared = .106)

Parameter Estimates

Dependent Variable: Distensibility

Parameter	B	Std. Error	t	Sig.	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound	Partial Eta Squared
Intercept	.004	.000	27.871	<.001	.004	.004	.282
PASE_centred	4.457E-7	3.041E-7	1.466	.143	-1.507E-7	1.042E-6	.001
[Sex_A=0]	-2.679E-5	3.973E-5	-.674	.500	.000	5.113E-5	.000
[Sex_A=1]	0[a]	.	.	.	.	.	.
Age	-2.990E-5	2.176E-6	-13.742	<.001	-3.417E-5	-2.563E-5	.087

a. This parameter is set to zero because it is redundant.

General Linear Model: Adjusted Model with No Interactions and No Outliers

Univariate Analysis of Variance

Between-Subjects Factors

		N
Sex/Gender	0	726
	1	997
Heart_disease_no_missing	1.00	296
	2.00	1427
Smoking_Status_1	1.00	716
	2.00	831
	3.00	176

Levene's Test of Equality of Error Variances[a]

Dependent Variable: Distensibility

F	df1	df2	Sig.
1.072	11	1711	.380

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.[a]

a. Design: Intercept + PASE_centred + Sex_A + Age + Heart_disease_no_missing + Smoking_Status_1 + SBP_centred + hsCRP_centred + cIMT_centred + HBA1c_centred + Trig_centred + HDL_centred + LDL_centred + DBP_centred + BMI_centred

Tests for Heteroskedasticity

White Test for Heteroskedasticity[a,b,c]

Chi-Square	df	Sig.
159.337	130	.041

a. Dependent variable: Distensibility
b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.
c. Design: Intercept + PASE_centred + Sex_A + Age + Heart_disease_no_missing + Smoking_Status_1 + SBP_centred + hsCRP_centred + cIMT_centred + HBA1c_centred + Trig_centred + HDL_centred + LDL_centred + DBP_centred + BMI_centred + PASE_centred * PASE_centred + Sex_A * PASE_centred + PASE_centred * Age + Heart_disease_no_missing * PASE_centred + Smoking_Status_1 * PASE_centred + PASE_centred * SBP_centred + PASE_centred * hsCRP_centred + PASE_centred * cIMT_centred + PASE_centred * HBA1c_centred + PASE_centred * Trig_centred + PASE_centred * HDL_centred + PASE_centred * LDL_centred + PASE_centred * DBP_centred + PASE_centred * BMI_centred + Sex_A * Age + Sex_A * Heart_disease_no_missing + Sex_A * Smoking_Status_1 + Sex_A * SBP_centred + Sex_A * hsCRP_centred + Sex_A * cIMT_centred + Sex_A * HBA1c_centred + Sex_A * Trig_centred + Sex_A * HDL_centred + Sex_A * LDL_centred + Sex_A * DBP_centred + Sex_A * BMI_centred + Age * Age + Heart_disease_no_missing * Age + Smoking_Status_1 * Age + Age * SBP_centred + Age * hsCRP_centred + Age * cIMT_centred + Age * HBA1c_centred + Age * Trig_centred + Age * HDL_centred + Age * LDL_centred + Age * DBP_centred + Age * BMI_centred + Heart_disease_no_missing * Smoking_Status_1 + Heart_disease_no_missing * SBP_centred + Heart_disease_no_missing * hsCRP_centred + Heart_disease_no_missing * cIMT_centred + Heart_disease_no_missing * HBA1c_centred + Heart_disease_no_missing * Trig_centred +

Heart_disease_no_missing * HDL_centred +
Heart_disease_no_missing * LDL_centred +
Heart_disease_no_missing * DBP_centred +
Heart_disease_no_missing * BMI_centred +
Smoking_Status_1 * SBP_centred +
Smoking_Status_1 * hsCRP_centred +
Smoking_Status_1 * cIMT_centred +
Smoking_Status_1 * HBA1c_centred +
Smoking_Status_1 * Trig_centred +
Smoking_Status_1 * HDL_centred +
Smoking_Status_1 * LDL_centred +
Smoking_Status_1 * DBP_centred +
Smoking_Status_1 * BMI_centred +
SBP_centred * SBP_centred + SBP_centred *
hsCRP_centred + SBP_centred * cIMT_centred
+ SBP_centred * HBA1c_centred + SBP_centred
* Trig_centred + SBP_centred * HDL_centred +
SBP_centred * LDL_centred + SBP_centred *
DBP_centred + SBP_centred * BMI_centred +
hsCRP_centred * hsCRP_centred +
hsCRP_centred * cIMT_centred +
hsCRP_centred * HBA1c_centred +
hsCRP_centred * Trig_centred + hsCRP_centred
* HDL_centred + hsCRP_centred *
LDL_centred + hsCRP_centred * DBP_centred +
hsCRP_centred * BMI_centred + cIMT_centred
* cIMT_centred + cIMT_centred *
HBA1c_centred + cIMT_centred * Trig_centred
+ cIMT_centred * HDL_centred + cIMT_centred
* LDL_centred + cIMT_centred * DBP_centred
+ cIMT_centred * BMI_centred +
HBA1c_centred * HBA1c_centred +
HBA1c_centred * Trig_centred +
HBA1c_centred * HDL_centred +
HBA1c_centred * LDL_centred +
HBA1c_centred * DBP_centred +
HBA1c_centred * BMI_centred + Trig_centred *
Trig_centred + Trig_centred * HDL_centred +
Trig_centred * LDL_centred + Trig_centred *
DBP_centred + Trig_centred * BMI_centred +
HDL_centred * HDL_centred + HDL_centred *
LDL_centred + HDL_centred * DBP_centred +
HDL_centred * BMI_centred + LDL_centred *
LDL_centred + LDL_centred * DBP_centred +
LDL_centred * BMI_centred + DBP_centred *

DBP_centred + DBP_centred * BMI_centred + BMI_centred * BMI_centred

Modified Breusch-Pagan Test for Heteroskedasticity[a,b,c]

Chi-Square	df	Sig.
52.940	1	<.001

a. Dependent variable: Distensibility
b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.
c. Predicted values from design: Intercept + PASE_centred + Sex_A + Age + Heart_disease_no_missing + Smoking_Status_1 + SBP_centred + hsCRP_centred + cIMT_centred + HBA1c_centred + Trig_centred + HDL_centred + LDL_centred + DBP_centred + BMI_centred

Breusch-Pagan Test for Heteroskedasticity[a,b,c]

Chi-Square	df	Sig.
58.035	1	<.001

a. Dependent variable: Distensibility
b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.
c. Predicted values from design: Intercept + PASE_centred + Sex_A + Age + Heart_disease_no_missing + Smoking_Status_1 + SBP_centred + hsCRP_centred + cIMT_centred + HBA1c_centred + Trig_centred + HDL_centred + LDL_centred + DBP_centred + BMI_centred

F Test for Heteroskedasticity[a,b,c]

F	df1	df2	Sig.
54.555	1	1721	<.001

a. Dependent variable: Distensibility
b. Tests the null hypothesis that the variance of the errors does not depend on the values of the independent variables.
c. Predicted values from design: Intercept + PASE_centred + Sex_A + Age + Heart_disease_no_missing + Smoking_Status_1 + SBP_centred + hsCRP_centred + cIMT_centred + HBA1c_centred + Trig_centred +

HDL_centred + LDL_centred + DBP_centred + BMI_centred

Tests of Between-Subjects Effects

Dependent Variable: Distensibility

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	.000[a]	15	2.379E-5	37.552	<.001	.248
Intercept	.000	1	.000	571.361	<.001	.251
PASE_centred	.000	1	.000	.000	1.000	.000
Sex_A	2.689E-6	1	2.689E-6	4.244	.040	.002
Age	9.611E-5	1	9.611E-5	151.701	<.001	.082
Heart_disease_no_missing	2.313E-6	1	2.313E-6	3.650	.056	.002
Smoking_Status_1	1.889E-6	2	9.445E-7	1.491	.226	.002
SBP_centred	3.825E-5	1	3.825E-5	60.375	<.001	.034
hsCRP_centred	1.794E-7	1	1.794E-7	.283	.595	.000
cIMT_centred	3.990E-6	1	3.990E-6	6.298	.012	.004
HBA1c_centred	1.164E-5	1	1.164E-5	18.366	<.001	.011
Trig_centred	8.142E-6	1	8.142E-6	12.851	<.001	.007
HDL_centred	1.060E-9	1	1.060E-9	.002	.967	.000
LDL_centred	1.101E-6	1	1.101E-6	1.738	.188	.001
DBP_centred	3.603E-6	1	3.603E-6	5.687	.017	.003
BMI_centred	7.247E-6	1	7.247E-6	11.439	<.001	.007
Error	.001	1707	6.336E-7			
Total	.009	1723				
Corrected Total	.001	1722				

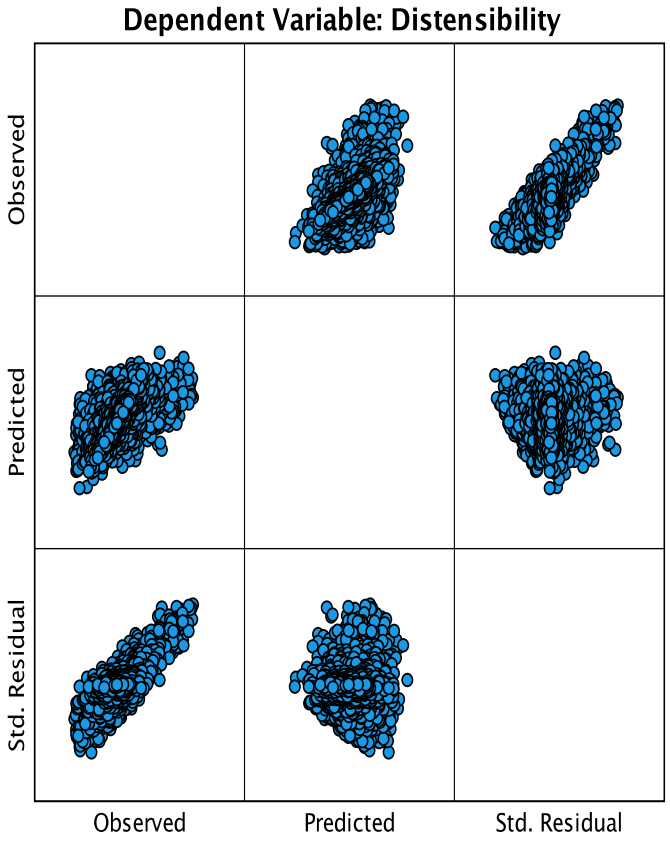
a. R Squared = .248 (Adjusted R Squared = .242)

Parameter Estimates

Dependent Variable: Distensibility

Parameter	B	Std. Error	t	Sig.	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound	Partial Eta Squared
Intercept	.004	.000	23.284	<.001	.004	.005	.241
PASE_centred	7.331E-10	3.053E-7	.002	.998	-5.981E-7	5.996E-7	.000
[Sex_A=0]	-9.370E-5	4.548E-5	-2.060	.040	.000	-4.492E-6	.002
[Sex_A=1]	0[a]	.	.	.	.	.	.
Age	-3.333E-5	2.706E-6	-12.317	<.001	-3.864E-5	-2.802E-5	.082
[Heart_disease_no_missing=1.00]	-9.765E-5	5.111E-5	-1.911	.056	.000	2.593E-6	.002
[Heart_disease_no_missing=2.00]	0[a]	.	.	.	.	.	.
[Smoking_Status_1=1.00]	8.810E-5	6.831E-5	1.290	.197	-4.587E-5	.000	.001
[Smoking_Status_1=2.00]	.000	6.806E-5	1.716	.086	-1.670E-5	.000	.002
[Smoking_Status_1=3.00]	0[a]	.	.	.	.	.	.
SBP_centred	-1.321E-5	1.700E-6	-7.770	<.001	-1.654E-5	-9.875E-6	.034
hsCRP_centred	-1.907E-6	3.584E-6	-.532	.595	-8.936E-6	5.122E-6	.000
cIMT_centred	.000	.000	2.510	.012	6.707E-5	.001	.004
HBA1c_centred	-8.063E-5	1.882E-5	-4.286	<.001	.000	-4.373E-5	.011
Trig_centred	-9.396E-5	2.621E-5	-3.585	<.001	.000	-4.255E-5	.007
HDL_centred	2.367E-6	5.788E-5	.041	.967	.000	.000	.000
LDL_centred	2.850E-5	2.162E-5	1.318	.188	-1.390E-5	7.090E-5	.001
DBP_centred	-6.857E-6	2.876E-6	-2.385	.017	-1.250E-5	-1.217E-6	.003
BMI_centred	-1.406E-5	4.156E-6	-3.382	<.001	-2.221E-5	-5.906E-6	.007

a. This parameter is set to zero because it is redundant.



Model: Intercept + PASE_centred + Sex_A + Age + Heart_disease_no_missing + Smoking_Status_1 + SBP_centred + hsCRP_centred + cIMT_centred + HBA1c_centred + Trig_centred + HDL_centred + LDL_centred + DBP_centred + BMI_centred