Correlation of cardiac risk factors with carotid intima-media thickness and radial intima-media thickness measurements

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Declaration of Independent Work

I, Fritz-Gerald van Schalkwyk, with ID number _____, declare that this research project submitted to the Central University of Technology in fulfilment of the requirements for a **Master's Degree in Health Sciences in Clinical Technology** is my independent work and has not been submitted to any other institution by myself or another individual in fulfilment of my degree.

June 2023

Signature **Date**

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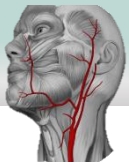
Symbols, SI Units and Abbreviations



&	And
>	Bigger than
=	Equals to
µ	Micro
%	Percentage
±	Plus/minus
<	Smaller than
ACS	Acute coronary syndrome
AF	Atrial fibrillation
AHA	American Heart Association
ANGPTL3	Angiopoietin-like 3
APOA-L	Apolipoprotein
BMI	Body mass index
BP	Blood pressure
CAD	Coronary artery disease
CIMT	Carotid intimal media thickness
CCA	Common carotid artery
CETP	Cholesteryl ester transfer protein
CM	Centimeter
CRP	C-reactive protein
CT	Computed tomography
CVA	Cerebrovascular accidents
CVD	Cardiovascular disease
ECA	External carotid artery
EC	Endothelial cells
ESR	Erythrocyte sedimentation rate
EPC	Epilepsia partialis continua
HDLC	High-density lipoprotein-cholesterol
HDL	High-density lipoprotein
HIV	Human Immunodeficiency Virus
HR	Heart rate

ICA	Internal carotid artery
IHD	Ischemic heart disease
IMT	Intimal media thickness
Kg/m²	Kilogram-Meter Squared
LAD	Left anterior descending
LDLC	Low-density lipoprotein cholesterol
LDL	Low-density lipoprotein
LP(a)	Lipoprotein(a)
LVH	Left ventricular hypertrophy
Mg/dl	Milligrams per decilitre
MI	Myocardial infarct
Mm	Millimeter
mmHg	Millimeter of mercury
mmol/L	Millimoles per litre
MRI	Magnetic resonance imaging
MACS	Multicenter AIDS Cohort study
NMR	Nuclear magnetic resonance
NO	Nitric oxide
RA	Rheumatoid arthritis
RIMT	Radial intimal media thickness
RHPAT	Reactive hyperaemia peripheral arterial tonometry
SA	Sinoatrial
SABPA	Sympathetic Activity and Ambulatory Blood Pressure in Africans
SD	Standard deviation
SMC	Smooth muscle cell
SV	Stroke volume
TCFA	Thin-cap fibroatheroma
TIA	Transient ischemic attacks
TRL	Toll-like receptors (TLR's)
VTE	Venous thromboembolism
TPR	Total peripheral resistance
VLDL	Very-low-density lipoprotein
WBC	White blood cells
WIHS	Womens Interagency HIV study

WSS	Wall shear stress

Glossary

All definitions in the glossary were obtained from Medical Terms and Abbreviations: Merriam-Webster Medical Dictionary [https://www.merriam-webster.com/]

TERM	DEFINITION
A	
Aneurysmal	An abnormal blood-filled bulge of a blood vessel and especially an artery resulting from weakening (as from disease) of the vessel wall.
Angiography	The radiographic visualization of the blood vessels after injection of a radiopaque substance.
Atherosclerosis	Atheromatous deposits in and fibrosis of the inner layer of the arteries.
C	
Cardiovascular disease (CVD)	Disease relating to or involving the heart and blood vessels.
Carotid artery	Either of the two main arteries that supply blood to the head of which, the left in humans arises from the arch of the aorta and the right by bifurcation of the brachiocephalic artery.
Clavicle	A bone of the shoulder girdle typically serving to link the scapula and sternum.
Computed tomography	Radiography in which a three-dimensional image of a body structure is constructed by a computer from a series of plane cross-sectional images made along an axis.
D	
Diabetes Mellitus (DM)	A variable disorder of carbohydrate metabolism caused by a combination of hereditary and environmental factors and usually characterized by inadequate secretion or utilization of insulin, by excessive urine production, by excessive

	amounts of sugar in the blood and urine, and by thirst, hunger, and loss of weight.
Doppler ultrasound	Ultrasound that utilizes the Doppler effect to measure movement or flow in the body and especially blood flow.
H	
Human Immunodeficiency Virus (HIV/AIDS)	Either of two retroviruses that infect and destroy helper T cells of the immune system causing the marked reduction in their numbers that is diagnostic of AIDS.
Hypertension	Abnormally high blood pressure and especially arterial blood pressure (systolic > 140 mmHg and diastolic > 95 mmHg).
I	
Infection	The state produced by the establishment of one or more pathogenic agents (such as bacteria, protozoans, or viruses) in or on the body.
M	
Macrophages	A phagocytic tissue cell of the immune system that may be fixed or freely motile; is derived from a monocyte, functions in the destruction of foreign antigens (such as bacteria and viruses), and serves as an antigen-presenting cell.
Magnetic resonance imaging	A non-invasive diagnostic technique that produces computerized images of internal body tissues and is based on nuclear magnetic resonance of atoms within the body induced by the application of radio waves.

Monocytes	A large white blood cell with finely granulated chromatin dispersed throughout the nucleus that is formed in the bone marrow enters the blood and migrates into the connective tissue, where it differentiates into a macrophage.
Myocardial Infarction (MI)	An acute episode of coronary heart disease marked by the death or damage of heart muscle due to insufficient blood supply to the heart, usually as a result of a coronary artery becoming blocked by a blood clot formed in response to a ruptured or torn fatty arterial deposit.
O	
Obesity	A condition characterized by the excessive accumulation and storage of fat in the body.
P	
Palma manus from the regio-antebrachii anterior	A thick superficial muscle on each side of the neck that arises by one head from the first segment of the sternum and by a second from the inner part of the clavicle, that inserts into the mastoid process and occipital bone and that acts especially to bend, rotate, flex, and extend the head.
Platelets	Blood that assists in blood clotting by adhering to other platelets and to damaged epithelium.
R	
Rheumatoid arthritis (RA)	A usually chronic autoimmune disease that is characterized especially by pain, stiffness, inflammation, swelling, and sometimes destruction of joints.
S	
Sudden cardiac death	Death occurring within minutes or hours following onset of acute symptoms of cardiac arrest resulting especially from an arrhythmia.

T	
T cells	Several lymphocytes (such as a helper T cell) that differentiate in the thymus, possess highly specific cell-surface antigen receptors, and include some that control the initiation or suppression of cell-mediated and humoral immunity (as by the regulation of T and B cell maturation and proliferation) and others that lyse antigen-bearing cells.
Thrombosis	The formation or presence of a blood clot within a blood vessel.
V	
Vasospasm	Sharp and often persistent contraction of a blood vessel reducing its lumen and blood flow.
W	
White blood cells (WBC)	Any of the blood cells that are colorless, lack hemoglobin, contain a nucleus, and include the lymphocytes, monocytes, neutrophils, eosinophils, and basophils.

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Executive Summary

Introduction

Early identification of patients at risk for future cardiovascular events is essential since addressing these early stages is more effective than treating advanced atherosclerotic vascular disease. The presence of multiple risk factors increases the development of atherosclerosis significantly. Stroke is the second most common cause of mortality and the third most common cause of disability. Globally, ultrasound has been the most common method of evaluating carotid atherosclerosis due to reasonably low-cost ultrasound tests, easy access to the carotid artery, and the fact that the equipment is readily available. Carotid intima-media thickness (CIMT) is an independent predictor for stroke, and CIMT is positively associated with the risk of stroke, cardiovascular disease, and coronary artery disease.

Methods

A prospective, descriptive-analytical single-centre study investigated the possible relationship between CIMT, radial intima-media thickness (RIMT), and modifiable and non-modifiable cardiac risk factors. This study was conducted at a private physician practice in Bloemfontein. Patients were referred from the Free State, Northern Cape, and Lesotho. The study sample included 250 1st time-visiting patients presenting with one or more modifiable cardiac risk factors. The following data was recorded, and examinations were performed:

i) Demographic and anthropometric data: age (years), sex, ethnicity, geographic location, height(cm), weight(kg), and body mass index (BMI) (kg/m²).
ii) Clinical data: modifiable and non-modifiable risk factors and family history.
iii) Ultrasound data: carotid and radial intimal medial thickness were recorded for each patient that consented to participate in the study.
iv) Other clinical data recorded from the patient's medical file included: human immunodeficiency virus (HIV) status and rheumatoid arthritis (RA).

Results

As expected, most patients came from the Free State (97.2%) and only 2.8% from other provinces. Most of these patients resided in the Mangaung Metropolitan district (85.6%). The

mean age of patients was 60.2 years, with a female predominance (63.6% vs 36.4%); most patients were Caucasian (76%). In the total study population, 42% (n=104) of patients presented with three or more modifiable risk factors, 39% (n=98) with two, and 48 patients (19%) with only one modifiable risk factor. Hypertension was the most common modifiable risk factor (89%), followed by obesity (66%). Diabetes Mellitus (DM) was present in 30% of patients, of which 55% presented with DM type 1 and 45% with DM type 2. The mean CIMT of the total group was 0.8±0.2 mm, and the maximum CIMT was 1.9 mm (right carotid artery) and 1.7 mm (left carotid artery). In men, the mean CIMT was 0.9±0.2 mm, and in women, 0.8±0.3 mm. Twenty-four percent of the study population presented with a stroke, and 51% of patients diagnosed with stroke or transient ischemic accident (TIA) had an abnormal CIMT. An abnormally thickened CIMT (≥0.9 mm) was observed in 107 (43%) of the study population with a mean of 1.1 mm±0.15.

Discussion

Age and male sex were the only non-modifiable risk factors contributing to the risk of an increase in mean CIMT: each year of age added 11% to the risk of having a thickened CIMT. Apart from demonstrating a risk for thicker CIMTs, significantly more men had an abnormal CIMT compared to the normal CIMT group indicating male sex as a significant risk factor.
Our results show that individual risk factors played a role in thickened CIMT. Hypertension, DM, hypercholesterolemia, and smoking significantly contributed to a thickened CIMT, whereas only hypercholesterolemia was associated with a thickened RIMT.

Risk factor combinations like DM and hypertension and hypertension and smoking significantly increase the risk of having thicker CIMT measurements. In contrast, the only combination associated with a significant contribution to a thicker mean RIMT was hypertension and obesity. The odds ratios of the combinations were markedly higher than isolated risk factors, most likely indicating the summative effects when individual risk factors occur in combination. However, it appears that having hypercholesterolemia and DM or hypercholesterolemia and obesity reduces the risk of having a thickened CIMT. The authors speculate that many of these patients were on statin treatment which may lend protection against an increased CIMT.

Conclusion

In conclusion, this study shows that male sex, increased age, hypertension, DM, hypercholesterolemia, and smoking significantly contributed to a thickened CIMT, whereas only hypercholesterolemia was associated with a thickened RIMT. Among all risk factors, hypertension had the most significant impact on the mean CIMT compared to the other modifiable risk factors. Risk factor combinations were also associated with a thickened CIMT and RIMT, and combinations appeared to add to the summative risk.

Chapter 1 - Introduction

1.1 Background

The most significant cause of premature mortality and disability worldwide is attributed to cardiovascular disease (CVD) (Kerola *et al.*, 2021). In 2015, CVD affected an estimated 422.7 million people and were responsible for an estimated 17.9 million deaths worldwide, 31% of all global deaths (Global Burden of Diseases, Injuries, and Risk Factor, 2015). CVD is not only seen in high-income countries but has grown disproportionally in both middle- and low-income countries. By 2030 it is predicted that 23 600 000 people will die from CVD annually (Song *et al.*, 2020).

Atherosclerosis is a primary contributor that causes significant cardiovascular mortality and morbidity and is often only detected when patients first experience an important cardiovascular event (Sakakura *et al.*, 2013). Atherosclerosis is a progressive, chronic, inflammatory disease with a long asymptomatic phase (Aziz, 2016). It is a significant disease process that leads to angina pectoris, ischemic stroke, myocardial infarction (MI), peripheral artery disease, and death (Herrington *et al.*, 2016). Several well-known risk factors contribute to the development of atherosclerosis (Nuraini, 2015). Non-modifiable risk factors include gender, age, family history, and ethnicity, while modifiable risk factors include hypercholesterolemia, diabetes mellitus, obesity, smoking, and hypertension (Ramrakha and Hill, 2012).

During the subclinical stage of CVD, atherosclerosis can be identified using intravascular ultrasonography, coronary and carotid angiography, computed tomography, magnetic resonance imaging (MRI), and B-mode ultrasonography (Mitchell *et al.*, 2018). According to Mitchell *et al*. (2018), carotid intima-media thickness (CIMT), carotid plaque presence, and radial intima-media thickness (RIMT) are used to assess the risk for CVD events. Individuals with a thickened CIMT and higher carotid plaque scores are associated with an increased risk of future CVD events.

In 2020, about 28% of individuals aged 30-79 had an abnormal CIMT of 1.0 mm and higher worldwide. About 21% (816 million) aged 30-79 years presented with a carotid plaque, and 1.5% presented with carotid stenosis (58 million). Carotid stenosis, plaque, and increased IMT

were more common in men and older individuals compared to women and younger individuals (Song *et al.*, 2020).

Noteworthy, imaging studies demonstrated that statin therapy could halt, slow down or even reverse the progression of atherosclerosis. Statin therapy reduces the atherogenic lipoprotein burden and is associated with significant changes in the rate of carotid and coronary atherosclerotic disease progression (Nezu *et al.*, 2016).

The burden of radial and carotid atherosclerosis in Africa remains an epidemiological blind spot because published data on radial and carotid load is limited. No published data exist for patients residing in central South Africa. Therefore, prospective studies are needed to investigate the correlation between non- and modifiable risk factors and radial and carotid intimal media thickness in a cohort of patients in central South Africa to predict cardiovascular events.

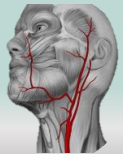
1.1.2 Aim

The study investigates the correlation between non- and modifiable risk factors and CIMT and RIMT to predict future cardiovascular events. In this study, the presence of atherosclerosis will be evaluated by examining the carotid and radial arteries.

1.1.3 Objectives

- Record demographic, anthropometric, and non- and modifiable risk factors.
- Measurement of the CIMT and RIMT in patients presenting with at least one modifiable cardiac risk factor visiting a private practice for the 1st time between November 2020–March 2021.
- Establish whether a correlation exists between risk factors and CIMT and RIMT.

Chapter 2 – Literature Review



2.1 Introduction

The leading cause of death and disabilities worldwide is CVD, including stroke and heart disease. CVD accounts for 17 million deaths annually (31% of total global deaths). More than half of the deaths occur before the age of 65. Premature deaths caused by coronary artery disease (CAD) are expected to increase by 41% in 2030 within the working group of the population (35-64 years) (Byrne *et al*., 2016). The second most common cause of death globally is stroke. From 1990 to 2017, the prevalence of stroke increased dramatically. By 2017, the number of people suffering from a stroke, primarily ischaemic stroke, was 104.2 million, which has doubled compared to 1990. The global rate of age-standardized stroke prevalence increased by 3% from 1990 to 2017 to 1300.6 per 100,000 in 2017 (Avan *et al.*, 2019). Notably, almost 90% of CVD, including myocardial infarction (MI) and stroke, is attributed to modifiable risk factors, including hypertension, hypercholesterolemia, DM, smoking, and obesity (Nezu *et al.*, 2016).

The burden of CVD has grown disproportionally in the past few decades, especially in low- and middle-income countries, where over 80% of CVD deaths occur (Mathers *et al*., 2006). In South Africa (SA), CVD is the leading cause of death. More South Africans die of CVD than all cancers combined (Byrne *et al*., 2016). Almost 1 in 6 deaths (17.3%) in South Africa are attributed to CVD, and 215 people die daily from strokes or heart disease. If 10 people have strokes and 5 have MI's, 10 people will die from these events every hour in South Africa (Hamer *et al.*, 2015). It is also important to note that there are growing inequalities in the occurrence and outcome of CVD between countries and social classes (Aziz *et al*., 2016).

According to Song *et al*. (2020), the primary pathological process contributing to CVD is atherosclerosis which can begin early in life while being asymptomatic and latent for long periods before progressing into advanced stages. Ultrasonography can be used for the early detection of atherosclerosis in healthy people by evaluating the carotid and peripheral arteries. CIMT can be determined using non-invasive ultrasound. According to the European Guidelines on CVD Prevention in Clinical Practice (2016), a CIMT of 1.0 mm or more is

considered abnormal (Song *et al.*, 2020). In individuals between 60-72 years, carotid calcifications were associated with future stroke events — the frequency of stroke increases with age. In the age group 78-96 years, the association between stroke and increased CIMT and carotid calcifications was insignificant (Bengtsson *et al.*, 2019). A few journals have reported a positive correlation between CVD and increased CIMT (Petty *et al.*, 1999 and O'Leary *et al.*, 1999) and the positive correlation between increased CIMT and the incidence of stroke amongst Caucasians (Harris, 2012). Harris (2012) observed a strong association between a thickened CIMT and stroke in the Indonesian population. The direct correlation exists because CIMT is a marker of generalized atherosclerosis. This pathologic vascular phenomenon plays a significant role in the pathogenesis of cardiovascular and cerebrovascular events such as stroke and defines the association between stroke and CIMT.

2.2 What is atherosclerosis?

Atherosclerosis is the most common type of arteriosclerosis, a disease of the large, medium, and small arteries characterized by the gradual build-up of fatty plaques, monocytes, platelets, and macrophages within the arterial wall (Lazenby, 2011). Atherosclerotic lesions are recognized as asymmetric focal thickenings of the intima, the innermost layer of the artery. Atherosclerosis causes a reduction in the vessel lumen diameter and eventually impairs blood flow to the distal tissues (Bentzon *et al.*, 2014). Acute coronary and carotid syndromes may also result from the formation of these plaques, which could trigger coronary and carotid thrombosis by becoming unstable (Ramrakha and Hill, 2012).

2.3 Location of atherosclerosis

Elastic and muscular arteries are most commonly affected by atherosclerosis, which often includes the aorta, carotid, coronary, and iliofemoral arteries. Arterial bifurcations and the proximal coronary arteries are more susceptible, and therefore are common sites for the development of atherosclerosis (Silva Marques and Pinto, 2014).

2.4 Pathophysiology of atherosclerosis

Atherosclerosis is a disease that is characterized by the accumulation of lipids, fibrous elements, and calcification within the large arteries. Endothelium activation initiates this development and is followed by a series of events, which implies activation of inflammatory pathways and vessel narrowing, leading to atheroma plaque formation. Altogether, these developments result

in cardiovascular complications that remain the leading cause of death worldwide (Jebari-Benslaiman *et al.*, 2022).

Atherosclerosis is a chronic inflammatory disease that begins with a fatty streak, a build-up of lipid-laden foam cells in the inner layer of the artery. Lipid retention is the beginning of the pathogenesis of atherosclerosis and is followed by chronic inflammation at susceptible sites in the major artery walls, which leads to fatty streaks and then progresses to fibroatheromas which are fibrous (Aziz, 2016) (Table 2.1). Leukocytes, endothelial- and intimal smooth muscle cells are the main contributors to the development of this condition (Falk, 2006).

As a result of endothelial dysfunction, atherosclerosis is initiated, accompanied by low-density lipoprotein (LDL) retention and modification of the intima. Atherogenic factors with modified LDL's promote the activation of endothelial cells leading to monocyte recruitment within the intima, which promotes foam cell formation (Jebari-Benslaiman *et al.*, 2022).

Oxygen-free radicals produced by the oxidation of fatty acids damage the artery, and endothelial cells initiate an immune and inflammatory reaction causing the attraction of monocytes, neutrophils, white blood cells (WBC), and platelets to the damaged area. Proinflammatory cytokines further aggravate the situation by attracting more WBC and platelets to the inflamed area, causing the activation of B- and T cells and stimulating clotting (Marieb and Hoehn, 2014).

The development of a fatty streak within an artery is an early indication of vessel damage, and platelet aggregation increases thrombus formation with continued inflammation and injury. The atheroma center (the core region) is formed by extracellular lipid droplets and foam cells, which are surrounded by a collagen-rich matrix and smooth muscle cells (Figure 2.1) (Marieb and Hoehn, 2014). Mast cells, T cells, and macrophages infiltrate the lesion and are plentiful in the shoulder region where the atheroma grows. The fatty streak consists primarily of macrophages and T cells. Atherosclerosis results in an increase in arterial stiffness and a decrease in vessel diameter (Jebari-Benslaiman *et al.*, 2022).

In addition, shear stress induces endothelial cell alignment (Figure 2.1a). In straight or tubular regions of the arteries where the wall shear stress (WSS) presents a laminar flow, endothelial cells show a flattened shape and an elongated alignment in the direction of the flow. At arterial bifurcations segments, flow disturbances occur and, as a consequence of the turbulent and reversal of flow, endothelial cells augment their appearance by adopting a cobblestone appearance (Figure 2.1a). Hemodynamic forces determine the early development of localised atherosclerotic plaque. Atherosclerotic lesions occur mainly in regions characterized by low WSS and flow separation (Figure 2.1b) and involve branch points and bifurcations (Jebari-Benslaiman *et al.*, 2022).

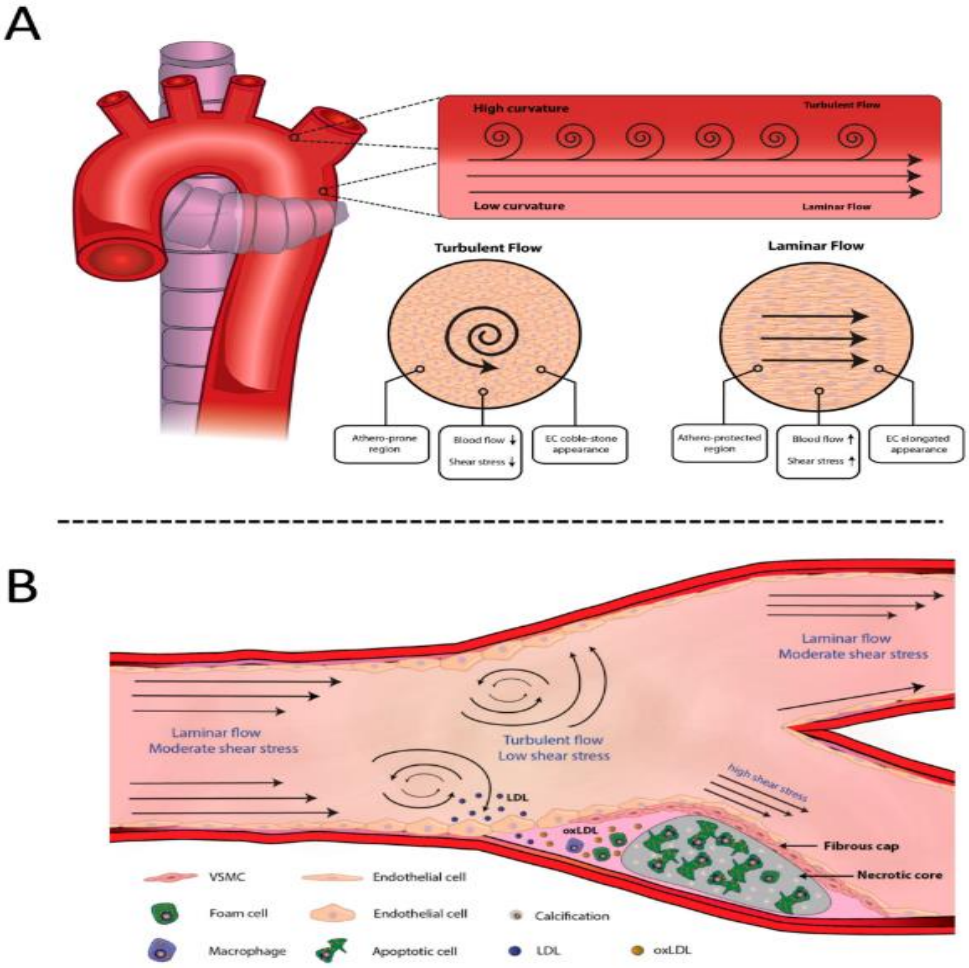


Figure 2.1. *Effect of flow and WSS patterns at arterial bifurcations on atherosclerotic plaque development* (Jebari-Benslaiman *et al.*, 2022). **A:** In straight vessel segments, physiological WSS with laminar flow leads to endothelial cells with a quiescent characteristic flattened shape. **B:** Turbulent flow occurs at bifurcations and branch points where the arterial curvature is higher due to flow separation.

2.5 Stages of atherosclerosis development

Atherosclerosis results from continuous progressive development, with the fatty streak developing at 11-12 years and fibrous plaques at 15-30 years (Aziz *et al.*, 2016). It is helpful to summarize lesion development into different classes of plaques. The later stages of lesion development are characterized by more advanced plaques identified by histopathology and appearance (Jebari-Benslaiman *et al.*, 2022). The advanced-stage plaques are responsible for clinical disease and are summarized in Table 2.1.

Table 2.1. The stages of atherosclerosis: Modified American Heart Association (AHA) consensus classification based on morphologic descriptions (Aziz, 2016)

Stage	Description	Thrombosis
	Nonatherosclerotic intimal lesions	
Intimal thickening	Normal accumulation of smooth muscle cells (SMCs) in the intima in the absence of lipid or macrophage foam cells.	Absent
Intimal xanthoma	Superficial accumulation of foam cells without a necrotic core or fibrous cap; based on animal and human data, such lesions usually regress.	Absent
	Progressive atherosclerotic lesions	
Pathologic intimal thickening	SMC-rich plaque with proteoglycan matrix and focal accumulation of extracellular lipid.	Absent
Fibrous cap atheroma	*Early necrosis:* focal macrophage infiltration into areas of lipid pools with an overlying fibrous cap. *Late necrosis:* loss of matrix and extensive cellular debris with an overlying fibrous cap.	Absent
Thin cap fibroatheroma	A thin, fibrous cap (<65µm) infiltrated by macrophages and lymphocytes with rare or absence of SMCs and a relatively large underlying necrotic core; intraplaque hemorrhage/ fibrin may be present.	Absent
	Lesions with acute thrombi	
Plaque rupture	Fibroatheroma with fibrous cap disruption; the luminal thrombus communicates with the underlying necrotic core.	Occlusive or non-occlusive
Plaque erosion	Plaque composition, as above; no communication of the thrombus with the necrotic core; can occur on a plaque substrate of pathologic intimal thickening or fibroatheroma.	Usually non-occlusive
Calcified nodule	Eruptive (shedding) of calcified nodules with an underlying fibrocalcific plaque with minimal or absence of necrosis.	Usually nonocclusive
	Lesions with healed thrombi	
Fibrotic (without calcification) **Fibrocalcific (± necrotic core)**	Collagen-rich plaque with significant luminal stenosis; lesions may contain large areas of calcification with few inflammatory cells and minimal or absence of necrosis; these lesions may represent healed erosions or ruptures.	Absent
Lesion reference to AHA types V and VI was discarded, because it failed to account for the 3 different morphologies (rupture, erosion, and calcified nodule) that give rise to acute thrombosis		

2.6 Clinical implications of atherosclerosis

Atherosclerosis gives rise to two types of plaques: stable and unstable (Bentzon *et al.*, 2014). Stable plaques regress, are static, or they grow slowly. Unstable plaques are complicated by erosion, fissure, and rupture and cause stenosis, thrombosis, and infarction. Most clinical events result from complications of unstable plaque. Therefore, plaque stabilization can reduce morbidity and mortality associated with atherosclerosis (Kajitani *et al.*, 2018).

Plaque rupture is caused by enzymes secreted by activating macrophages in the plaque. Once the plaque ruptures and the contents get exposed to circulating blood, the result is thrombosis (Bentzon *et al.*, 2014). The resultant thrombosis may initiate a change in plaque shape, occlude the lumen of a blood vessel, or the plaque contents may embolise. Low-risk or stable plaques are more fibrous and have a low lipid percentage, therefore do not cause 100% obstruction, while unstable plaques have a thick lipid core and thin fibrous cap narrow lumen (Aziz, 2016) (Figure 2.2).

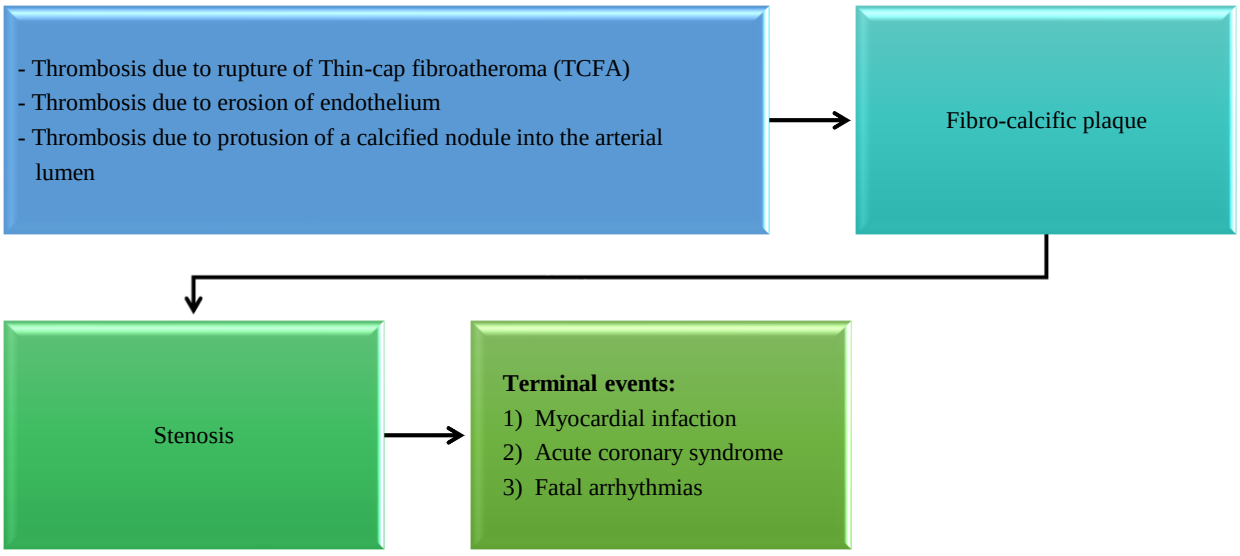


Figure 2.2. *Terminal events arise due to plaques and stenosis* (Aziz, 2016).

Sudden cardiac death and acute coronary syndrome (ACS) are related to four major classes of plaques. Arterial vessel damage occurs when plaques become fragile and rupture. This can cause the formation of blood clots that can break off and travel to another part of the body, or it can obstruct blood flow. If a clot obstructs a blood vessel that supplies the brain with blood,

it can initiate an ischemic stroke, and if the clot obstructs a blood vessel supplying the heart, it can cause a myocardial infarction (Figure 2.2) (Aziz, 2016).

Atherosclerosis is the cause of carotid artery disease or hardening of the arteries. The blockage or narrowing of the carotid arteries can prevent blood from reaching parts of the brain. A lack of blood causes oxygen shortages and can result in transient ischemic attacks (TIAs) or strokes (Iannuzzi *et al.*, 2021).

2.6.1 Two types of ischemic strokes

- A *thrombotic stroke* occurs when blood flow in a cerebral artery is compromised. An individual will experience one or more TIAs before a true thrombotic stroke occurs. A TIA is a short-lived, reversible disturbance in brain function resulting from cerebral hypoxia, and frequent TIAs suggest a true thrombotic stroke (Bekwelem *et al.*, 2016).
- An *embolic stroke* develops after arterial occlusion by an embolus formed outside the brain. Emboli leading to stroke include emboli breaking off from the common carotid arteries, atrial fibrillation (AF), or after a MI (Lazenby, 2011).

2.7 Atherosclerosis in the carotid and radial artery

The peripheral and carotid arteries have mainly been the focus for the early detection of atherosclerosis in apparently healthy individuals. The 2016 European Guidelines on Cardiovascular Disease Prevention in Clinical Practice stated that a CIMT of 1.0 mm or more is considered abnormal (Piepoli *et al.*, 2016). The Mannhein Carotid Intima-Media Thickness Consensus suggested that advanced atherosclerosis stages, including plaque, stenosis, and occlusion, are all indicators of cardiovascular risk (Aziz, 2016). Nezu *et al. (2016)* reported the global prevalence of moderate (≥50% stenosis) and severe (≥70% stenosis) carotid stenosis in asymptomatic individuals. However, there are no up-to-date estimates of the prevalence of carotid- and radial atherosclerosis globally. The availability of these statistics is imperative to understand the worldwide burden of carotid- and radial artery atherosclerosis to develop effective primary prevention and management strategies and inform relevant stakeholders.

2.8 Risk factors for the development of atherosclerosis

Nezu *et al.* (2016) reported that increased CIMT is associated with ageing, vascular risk factors, and the prevalence of CVD. Raitakari *et al.* (2003) reported that cardiovascular risk factors are correlated with increased CIMT. These cardiovascular risk factors include non-modifiable

(age, ethnic origin, family history, and gender) and modifiable risk factors (hypertension, DM, hypercholesterolemia, obesity, and smoking) (Aziz, 2016). However, other risk factors besides non- and modifiable risk factors have also been linked to the development of atherosclerosis and a thickened IMT. These risk factors include but are not limited to inflammation, sleep apnea, infection, HIV, and alcohol (Ren *et al.*, 2015).

A systemic review, meta-analysis, and modeling study published in The Lancet Global Health by Song *et al*. (2020) reported the prevalence and risk factors of carotid atherosclerosis using increased CIMT, carotid plaque, and stenosis in the general population. They noted that smoking, diabetes, and hypertension were significant risk factors for increased CIMT and carotid plaque. Advanced age was a risk factor for carotid stenosis. According to Rockman *et al*. (2013), racial differences must also be considered when evaluating the prevalence of carotid atherosclerosis. The research group reported that Native American and Caucasian individuals had the highest prevalence of carotid stenosis, whereas African American males and Asian females appeared to have the lowest prevalence. Therefore, racial differences as a risk factor for the prevalence of carotid stenosis might be associated with the frequency of atherosclerotic risk factors in various races.

2.8.1 Non-modifiable risk factors

Non-modifiable risk factors cannot be controlled, including a family history of cardiovascular disease, age, sex, and ethnicity (Boehme *et al*., 2017).

2.8.1.1 Family history of cardiovascular disease

A family history of ischemic heart disease (IHD) is an independent and significant risk factor for CAD (Shea *et al.*, 1984). According to Atkins *et al.* (2019), patients with a family history of CVD demonstrated a 77% higher total carotid plaque burden than those with no family history of CVD. Therefore, the total carotid plaque area is associated with a family history of CVD.

A positive family history of CVD is associated with an 84% increase in women and a 75% increase in men to develop CVD, or more than double if both parents are affected (Ramrakha and Hill, 2012). For this reason, carotid ultrasound screening in patients with a family history of CVD is recommended, as these patients are considered at high risk for developing CVD and

can benefit from early detection and intervention. Atherosclerosis is a polygenic, multifactorial disorder caused by interactions between the effects of variations in the genetic sequence of several genes and lifestyles (Bentzon *et al.*, 2014). Family history is considered essential when the atherosclerotic disease presents in a first-degree female relative before 65 years and/or male relative before 55 years, and the risk of atherosclerosis doubles when both parents are affected with atherosclerosis (Ramrakha and Hill, 2012).

2.8.1.2 Age

A significant risk factor for atherosclerotic disease is ageing because of the degenerative process associated with ageing and the impact of the worsening risk factor profile that develops with increased age (Herrington *et al.*, 2016). CAD causes over 40 000 premature deaths (<75 years), of which 22% is seen in men and 13% in women (Boehme *et al*., 2017). Fifteen percent of men and 9% of women have symptomatic CAD by the age of 70, and by the age of 80, it increases to 20%. In people older than 65 years, 80% of deaths are attributed to MI. MI occurs in 45% of people under 65, but in older individuals, the MI is more likely to be fatal (Aziz, 2016).

The process of vascular ageing already starts in childhood. Arterial ageing may primarily be viewed from two viewpoints. Firstly, it involves the process of arterial stiffening and loss of elasticity. Secondly, degenerative changes and the formation of atherosclerotic plaques occur, this being the cause of ischemia. By the age of 70 years, 15% of men and 9% of women have symptomatic CAD increasing to 20 % by the age of 80 years (Petrák and Češka, 2019).

2.8.1.3 Gender

CAD is more common in men, and the onset of CAD also tends to be earlier in men than in women (Ramrakha and Hill, 2012). In the United States of America, CVD is the leading cause of death for men and women (Villablanca *et al.*, 2010). According to the American Heart Association (2012), mortality in women due to CVD outpaces men because;

- women have higher death rates following MI or stroke,
- higher prevalence of cardiac risk factors and cardiovascular events
- lower awareness of CVD.

Estrogen is generally believed to protect women from CVD in the premenopausal age group (Mathur *et al.*, 2015). Interestingly, a preponderance of vascular and connective tissue

disorders in women also points to an inherent role of hormones and tissue factors in maintaining vascular endothelial function. The differential effect of lipoprotein A, toll-like receptors (TLRs), and leucocyte-platelet aggregate markers in women and men also suggests inherent gender-related differences in the pathophysiology of atherosclerosis. A better understanding of the pathophysiology will likely open ways to improve evidence-based treatment of CVD in women (Mathur *et al.*, 2015).

2.8.1.4 Ethnic origin

Between ethnic groups, the burden of atherosclerotic disease varies (Banerjee and Chimowitz, 2017). Cardiovascular mortality is more significant in blacks than in whites (Avan *et al.*, 2019). Bennet *et al*. (2010) reported a greater maximum CIMT in Blacks than South Asians. The 3-year follow-up Sympathetic Activity and Ambulatory Blood Pressure in Africans (SABPA) study demonstrated a similar prevalence of carotid IMT stenosis among black Africans compared to whites. However, in black subjects, they noted an increase in the number of cardiac risk factors compared to whites (Hamer *et al.*, 2015). Rockman *et al.* (2013) also reported that the incidence of stroke is higher in blacks than whites.

Judd *et al.* (2013) reported that African Americans are ± 50% more likely to have a stroke than their white adult counterparts, and black men are 70% more likely to die than black non-Hispanic white men. High blood pressure is present in over 50% of black adults and develops earlier. More than 60% of black men and 80% of black women are overweight or obese in South Africa, and more African populations have high levels of LDL and are diagnosed with DM (Muller *et al.*, 2019). Over 20% of the black adult population are smokers, doubling their risk of stroke (Kondo *et al.*, 2019).

2.8.2 Modifiable CVD risk factors

Nezu *et al.* (2016) stated that several risk factors are associated with increased IMT; (i) age, (ii) hypertension, (iii) cholesterol, (iv) smoking, and (v) DM. Modifiable risk factors are defined as risk factors that can be controlled (Hajar *et al*., 2017) and are summarized in Figure 2.3.

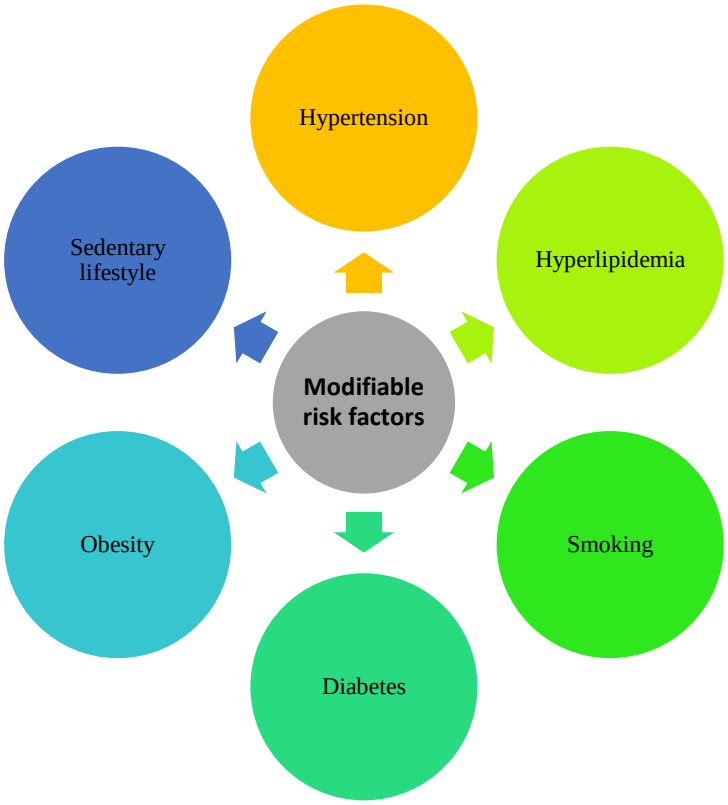


Figure 2.3. *Modifiable cardiovascular risk factors* (Herrington *et al.*, 2016).

This study evaluated modifiable CVD risk factors, as the correlation between CIMT and RIMT and modifiable CVD risk factors was investigated.

2.8.2.1 Hypertension

Chronically high blood pressure produces a shear force that scrapes away at the endothelial layer of the arteries initiating injury. Shear forces mainly occur at sites of arterial bifurcation, including the aorta, cerebral- and carotid arteries (Marieb and Hoehn, 2014). In 2010 approximately 1.39 billion adults suffered from hypertension worldwide (31.1%). The prevalence in lower-income countries (31.5%, 1.04 billion people) is higher when compared to high-income countries (28.5%, 349 million people) (Mills *et al.*, 2020).

Shrestha *et al.* (2009) reported that an increase in systolic blood pressure correlates with atherosclerotic changes visible in the carotid arteries and with high and low-grade lesions. Furthermore, hypertensive patients with cognitive dysfunction demonstrate changes in their vascular morphology, characterized by an increased carotid IMT, impaired hemodynamic function, and enhanced atherosclerotic lipid profile, manifested by an elevated central systolic blood pressure (Dias *et al.*, 2012).

A continuous relationship exists between cardiovascular risk and elevated blood pressure (Ramrakha and Hill, 2012). Hypertension is the most prevalent risk factor for CVD. Therefore, the higher the blood pressure, the higher the risk for CVD (Lazenby, 2011). Atherosclerotic coronary arteries are stiff and cannot dilate to supply adequate amounts of oxygen to the myocardium and can cause a MI. Long-standing hypertension can also cause left ventricular hypertrophy, which initiates changes in the heart's conduction system leading to dysrhythmia, increased risk of clot formation, and myocardial ischemia leading to a MI (Lazenby, 2011).

Hypertension is classified according to systolic and diastolic pressure, and a patient is considered hypertensive if the systolic blood pressure exceeds 139 mmHg and the diastolic blood pressure exceeds 89 mmHg (Nuraini, 2015).

Table 2.2. Classification of hypertension (Ramrakha and Hill, 2012)

Category	Systolic blood pressure (mmHg)	Diastolic blood pressure (mmHg)
Normal BP	<130	<85
High-normal BP	130-139	85-89
Grade 1 hypertension	140-159	90-99
Grade 2 hypertension	160-179	100-109
Grade 3 hypertension	≥180	≥110

2.8.2.2 Hypercholesteremia

Cholesterol is a waxy, soft substance found in all the body cells and the bloodstream (Kayashima and Maeda-Smithies, 2020). The most common fat in the body is triglycerides and high levels of low-density-lipoproteins (LDL). Cholesterol and triglycerides speed up atherosclerosis and increase the risk of stroke (Marieb and Hoehn, 2014). Lipoproteins transport high-density-lipoproteins (HDL) and LDL cholesterol through the blood. HDL is known to be protective against atherosclerosis and CVD because HDL carries fat away from cells to be degraded. LDL and very low-density lipoprotein (VLDL) carry fat to the body's cells, including the endothelial cells of the arteries (Lazenby, 2011). High levels of triglycerides and high serum cholesterol can cause the development of atherosclerosis (Naomi *et al.*, 2013). Atheromas are found inside and throughout the tunica media of arteries in individuals with atherosclerosis (Toth, 2008). The normal ranges for plasma lipid levels are summarized in Table 2.3.

Table 2.3. Normal ranges for plasma lipid levels (Ramrakha and Hill, 2012)

Parameter	Reference range (mg/dL)	Reference range (mmol/L)
LDL cholesterol	<160 mg/dL	<4.1 mmol/L
HDL cholesterol	<30-75 mg/dL	0.8-2.0 mmol/L
Total cholesterol	150-250mg/dL	4.0-6.5 mmol/L
Triglycerides	70-175 mg/dL	0.8-2.0 mmol/L

[LDL, low-density lipoproteins, HDL, high-density lipoproteins]

It is important to note that a patient's cholesterol value can be affected by several modifiable and non-modifiable risk factors (Kayashima and Maeda-Smithies, 2020), as summarized in Figure 2.4.

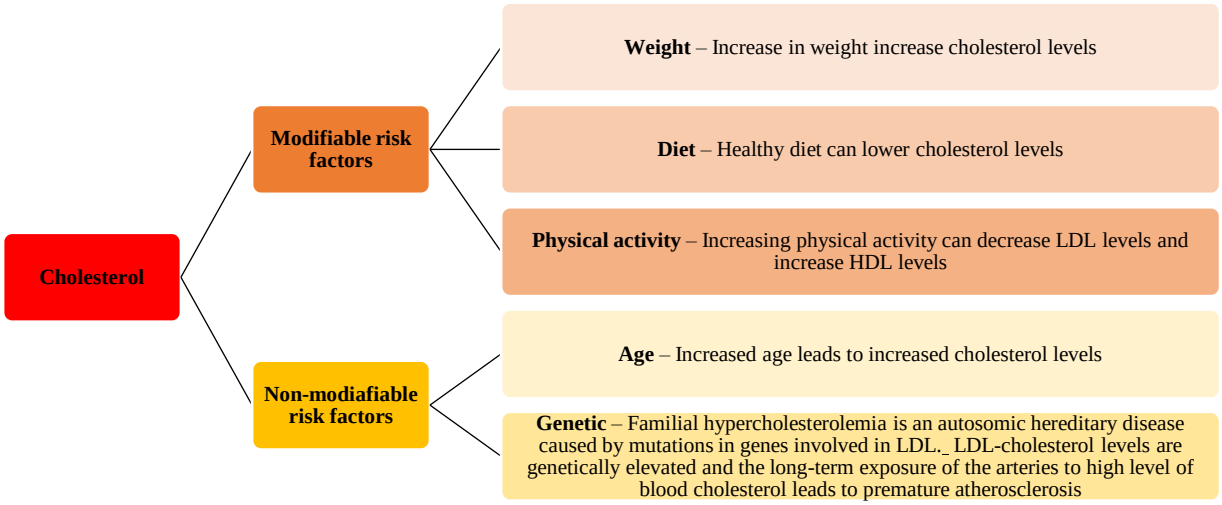


Figure 2.4. *Modifiable and non-modifiable risk factors influencing cholesterol values* (Bruckert and Gallo, 2017)**.**

2.8.2.3 Smoking

Smoking is an established risk factor for the development of atherosclerosis (Kondo *et al.*, 2019). Both active and passive smoking can contribute to the development and progression of atherosclerosis (Pan *et al.*, 2019). According to the Czech Guidelines for Tobacco Addiction Treatment (2010), smoking is the cause of 1 in every five deaths throughout Europe, resulting in 18 000 deaths annually (Kondo *et al.*, 2019). The risk of CAD increases by 50% due to smoking, and the mortality rate is 60% higher in smokers than non-smokers (Ramrakha and Hill, 2012).

Smoking tobacco products promotes thrombus formation and increases the risk of carotid and coronary plaque rupture (Kondo *et al.*, 2019). A healthy endothelium produces vasodilatory substances. However, the bioactivity and synthesis of the vasodilation process are impaired when the endothelium is injured, causing an imbalance between vasoconstrictors and vasodilators (Ramrakha and Hill, 2012). Cigarette smoke causes the upregulation of inflammatory cytokines and oxidative stress, causing endothelial dysfunction by reducing nitric oxide (NO) bioavailability (Herrington *et al.*, 2016).

Oxidation of LDL is caused by the formation of peroxynitrite and inflammatory cytokines, which enhance the atherosclerotic process causing endothelial dysfunction. The impairment of anticoagulative fibrinolysis, the coagulation cascade, and platelet activation are initiated by cigarette smoking (Kondo *et al.*, 2019). Figure 2.5 illustrates the mechanisms and pathways by which cigarette smoking causes CVD.

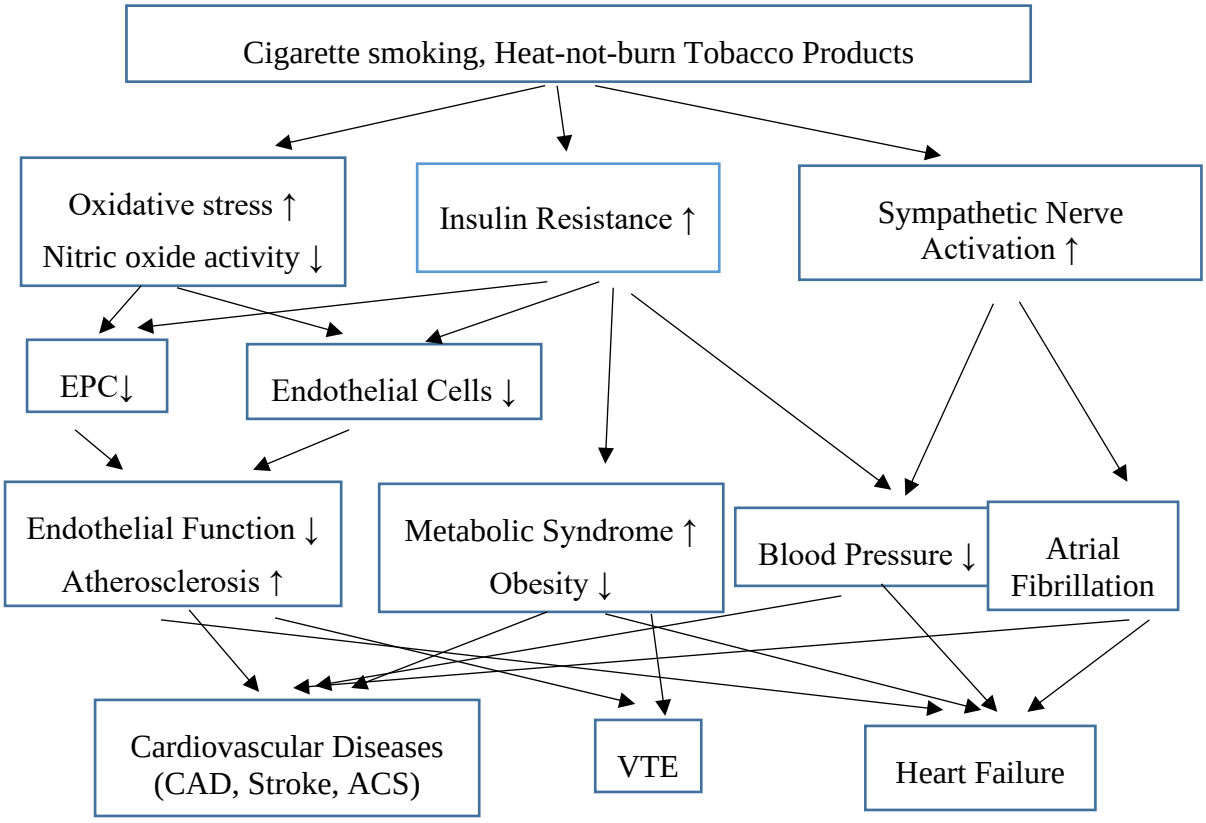


Figure 2.5. *Mechanisms and pathways by which cigarette smoking cause CVD***,** adapted from **(**Kondo *et al.*, 2019**).** [ECP, Epilepsia partialis continua, VTE, Venousthrombolism]**.**

2.8.2.4 Obesity

Central obesity is the accumulation of excess fat concentrated mainly in the abdomen, confirmed by a high waist-to-hip ratio (Nezu *et al.*, 2016). A multifactorial chronic disease characterized by an accumulation of subcutaneous and visceral fat is referred to as obesity, which leads to cardiometabolic diseases (Lovren *et al.*, 2015). Insulin resistance, lipid metabolism, endothelial dysfunction, adipokine imbalance, and inflammation have all been suggested to contribute to the underlying relationship between atherosclerosis and obesity (Ragino *et al.*, 2020). Obesity confers to a relatively high risk for atherosclerosis development, especially if the waist circumference is >88 cm in women and >102 cm in men (Song *et al.*, 2020). The height or weight of an individual does reflect on body composition. Therefore, to determine the degree of obesity, the body mass index (BMI) must be calculated (Marieb and Hoehn, 2014). BMI is the weight ratio in kilograms divided by the square of the height in meters (Bohlen *et al.*, 2015). Table 2.5 summarize obesity classifications according to BMI.

Table 2.4. Classification of BMI (World Health Organization, 2011)

Classification	BMI (kg/m^2)
Underweight	<18.5
Normal range	18.5-24.9
Overweight	25-29.9
Grade I obesity	30.0-34.9
Grade II obesity	35.0-39.9
Grade III obesity	>39.9

2.8.2.5 Diabetes mellitus (DM)

Diabetes mellitus (DM) is a chronic metabolic disorder characterized by persistent hyperglycemia. It may be due to resistance to peripheral actions of insulin or impaired insulin secretion, or both. Chronic hyperglycemia, in cooperation with the other metabolic abnormalities in patients with DM, can cause damage to various organ systems, leading to disabling and life-threatening health complications. The most significant complications are microvascular (retinopathy, nephropathy, and neuropathy) and macrovascular complications leading to a 2-fold to 4-fold increased risk of cardiovascular diseases (Atkins *et al.*, 2019). Atherosclerosis, diabetic macroangiopathy secondary to DM, causes cerebro-cardiovascular diseases, the leading cause of death in patients with DM, significantly reducing their quality of life. Alterations in vascular homeostasis due to endothelial and vascular smooth muscle cell

dysfunction are the main features of diabetic macroangiopathy. Prolonged exposure to hyperglycemia and insulin resistance, in combination with other risk factors such as obesity, arterial hypertension, and dyslipidemia, are significant contributors (Katakami, 2018). Increased formation of advanced glycation end-products (AGEs) and activation of the AGEs-receptor for the AGE axis, overproduction of reactive oxygen species, polyol and hexosamine flux, protein kinase C activation, and chronic vascular inflammation are considered critical factors in the development of atherosclerosis in patients with DM (Du *et al.*, 2013).

Long-term diabetes causes vascular abnormalities, which are responsible for the development of stroke, CAD, and peripheral vascular disease. The leading cause of mortality and morbidity in diabetic patients is CAD (Nezu *et al.*, 2016). Stroke is a common complication linked to especially type 2 diabetes due to the combined risk of hypertension and atherosclerosis (Lazenby, 2011).

Type I diabetic patients without a previous history of cardiovascular disease, the presence of advanced diabetic retinopathy is associated with a higher atherosclerotic burden in the carotid arteries. Carbonell *et al*. (2018) reported that the IMT was increased in children and adults with type II diabetes compared to sex- and age-matched healthy control subjects. Type I diabetic patients presented with more carotid plaques and a thickened IMT than nondiabetic individuals.

2.8.2.6 Sedentary lifestyle

Being inactive can lead to slow blood movement, which allows fatty materials to build up in the blood vessels, contributing to heart disease and stroke. The body's ability to process fats is decreased when inactive. Lipoprotein lipase is a vital enzyme the body produces to break down fat in the blood. When sitting, the body's production of lipoprotein lipase drops by about 90%, which makes it very difficult for the body to use fat. When the body doesn't use fat, the fat gets stored, making a patient susceptible to heart disease and stroke. Therefore, a sedentary lifestyle should be avoided to decrease the risk of CVD by indulging in daily exercise and a healthy diet (Lechner *et al.*, 2020).

2.8.3 Other risk factors related to the development of atherosclerosis

It is important to note that many other risk factors can also contribute to atherosclerosis development, including HIV, rheumatoid arthritis (RA), inflammatory diseases, and an

unhealthy diet (Scarno *et al.*, 2014). This study also evaluated whether RA contributed to a thickened IMT.

2.8.3.1 Rheumatoid arthritis (RA)

RA is a systemic autoimmune disease characterized by inflammatory arthritis and extra-articular involvement (Kerola *et al*., 2021). A few similarities shared between atherosclerosis, and RA are mast cell and T-cell activation and the production of pro-inflammatory cytokines (Scarno *et al.*, 2014). In RA patients, the C-reactive protein (CRP) is elevated, caused by the decrease in endothelial nitric oxide synthase, thus contributing to endothelial dysfunction (Kerola *et al*., 2021).

Cardiovascular death is more common among RA patients with an elevated erythrocyte sedimentation rate (ESR) than patients with a non-elevated ESR. If the inflammatory markers increase, the carotid artery IMT increases in healthy and RA patients. RA patients present with more arterial atherosclerotic plaque, and the carotid plaque correlates with the systemic inflammatory radiological index and disease duration. Abnormal vascular function occurs in RA patients and causes an acceleration in the atherosclerotic process (Komici *et al.*, 2021). The atherosclerotic process seems to be accelerated and includes:

- an impaired endothelial response,
- reduced arterial elasticity,
- increased arterial stiffness, and
- increased IMT.

For this reason, the early detection of subclinical atherosclerosis in RA patients is significant, because it can prevent disability, cardiovascular disease, and, ultimately, death (Scarno *et al.*, 2014).

2.9 Diagnosis of atherosclerosis

Modern advances in medical technology have equipped us with several diagnostic techniques that can be used to confirm the diagnosis of atherosclerosis. Some of the primary diagnostic tools utilized today include:

- Nuclear magnetic resonance (NMR) spectroscopy of lipo-profiles. This test identifies the amount of cholesterol carried by each LDL particle and indicates the size of the particle. NMR is recommended for high-risk individuals (Lazenby, 2011).

- Reactive hyperemia peripheral arterial tonometry (RH-PAT) is a non-invasive technique to identify individuals with early-stage atherosclerosis (Aziz, 2016).
- Imaging of the arteries may allow visualization of atherosclerotic lesions, and these tests include ultrasound, computed tomography (CT) and magnetic resonance imaging (MRI) (Shrestha *et al.*, 2009).
- Increased C-reactive protein (CRP) levels (Nezu *et al.*, 2016).
- CIMT assessment using ultrasound (Ramrakha and Hill, 2012).

In this study, CIMT was the diagnostic tool of choice to evaluate atheroma load thickness using non-invasive ultrasound imaging. Duplex and colour ultrasound combine structural and functional data, quantifying the degree of stenosis caused by atherosclerotic plaque, especially in superficial vessels, such as the carotid and limb arteries (Song *et al.*, 2020). It is often used as the first-line imaging modality because it is non-invasive, radiation-free, and portable. In superficial vessels, the high resolution of ultrasound can differentiate between components of the arterial wall. For instance, an early subclinical sign of plaque formation is the presence of pathological proliferation of the intimal-medial layer. Detecting this thickening can be used as a "window" to evaluate an individual's global cardiovascular health status (Syed *et al.*, 2019). This imaging technique also allows for assessing hemodynamic and morphologic abnormalities before making therapeutic decisions (Nezu *et al.*, 2016). As a single examination strategy, duplex sonography is optimal for a final diagnosis and treatment plan for patients with symptomatic carotid artery and radial artery stenosis. Many clinical practices perform surgery only based on duplex ultrasound results (Mitchell *et al.*, 2018).

2.9.1 Carotid ultrasound

Carotid ultrasound is used to measure CIMT and was first described by Pignoli *et al*. (1984) and validated by the same authors in 1986. A strong correlation was documented between measured arterial wall thickness using ultrasound and histological findings (Betriu-Bars and Fernández-Giráldez, 2012).

Carotid ultrasound strongly predicts overall vascular health, especially atheromatous plaques and CIMT (Table 2.6). Measured CIMT using ultrasound is an accepted marker to indicate cardiovascular disease (Nezu *et al.*, 2016). According to Song *et al.* (2020), 28% of individuals aged between 30-79 years had an abnormal CIMT of >0.9 mm, translating to over one billion

people presenting with at least one risk factor. Twenty-one percent of these patients had a carotid plaque (equivalent to approximately 816 million people). According to the Global Burden of Diseases, Injuries and Risk Factor Study (2015), 422.7 million people were affected by CVD, including stroke, which caused an estimated 17.9 million deaths worldwide, comprising 31% of global deaths. Increased CIMT is more common in older people and men than in younger people and women. In 2015, a third of global cases of thickened CIMT were located in the Western Pacific region. In contrast, the African region had the smallest share of increased CIMT cases, highlighting substantial variations in prevalence worldwide (Song *et al.*, 2020).

Carotid ultrasound has a few advantages compared to all the previously described methods, for this technique for early atherosclerotic diagnosis is not generalized (Song *et al.*, 2020). Repeatability and non-invasiveness make IMT measurements an attractive biomarker and a potentially helpful therapeutic marker in patients at increased cardiovascular risk (Nezu *et al.*, 2016).

2.9.1.1 Measurement of carotid IMT

The carotid IMT can be measured between the intimal-luminal and the medial-adventitial interfaces (Figure 2.5). The appropriate depth of focus ranges from 30–40 mm, using at least a 7 MHz linear array transducer (Nezu *et al.*, 2016).

The common carotid artery (CCA) should be visualized by placing the probe above the clavicle in a mediocranial direction. From this view, the sonographer must move the probe till the carotid bulbus is visible. The color mode can be initiated to identify blood flow velocity patterns in the frequency of amplitude mode. The pulsed wave Doppler should be placed within the lumen, and a Doppler spectrum can be documented after the angle has been optimized *(Hennerici et al., 2006)*. The probe should be placed to visualize the external carotid artery (ECA) lateral to the proximal part of the sternocleidomastoid muscle and above the clavicle. The internal carotid artery (ICA) can be visualized by tilting the transducer. Scanning of the common carotid artery (CCA), ICA, and ECA should commence in a standardized sequence, and the flow- and anatomic relationships can be obtained using cross- and longitudinal sections; this sequence must be used to differentiate between artefact and carotid lesions (Figure 2.6). The ICA can be distinguished from the ECA when a blood vessel branching off the ECA is visible. The ECA usually shows higher pulsatility in comparison to the ICA. The ICA blood

flow is usually noticeably slower than the CCA's (Song *et al.*, 2020). Once the carotid bulbus is located, the IMT must be measured 10 mm before the carotid bulbus (Loizou *et al.*, 2015).

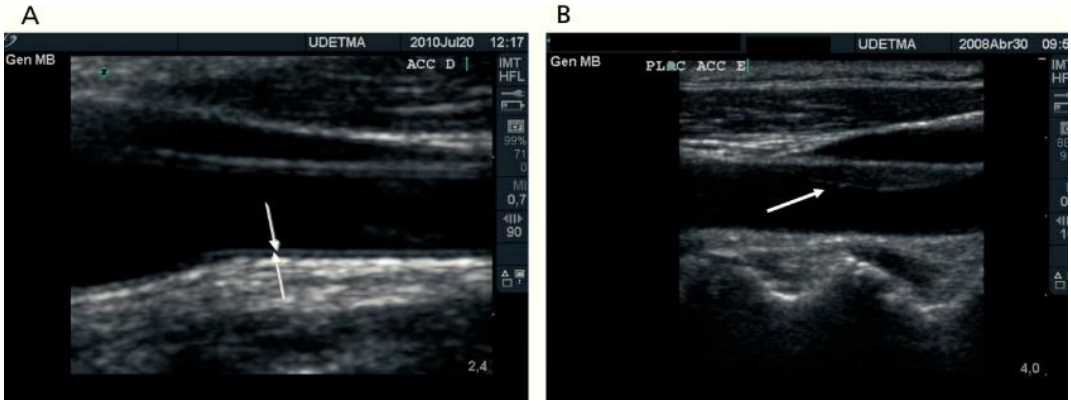


Figure 2.6. *Ultrasound image of carotid arteries* (Betriu-Bars and Fernández-Giráldez, 2012). **A:** View of the arterial wall for measuring the CIMT at the level of the common carotid artery. Arrows delimit the CIMT. **B:** Plaque along the anterior wall of the common carotid artery.

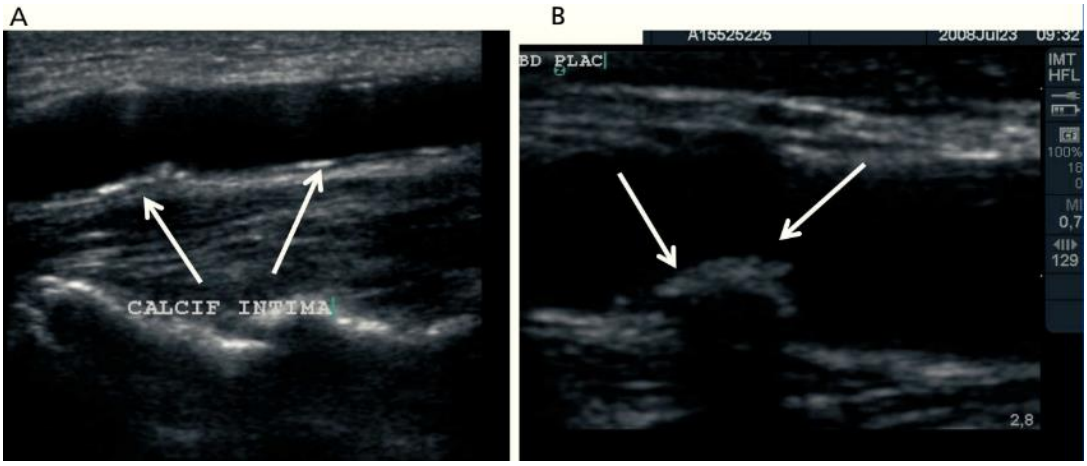


Figure 2.7. *Calcification patterns portrayed by ultrasound* (Betriu-Bars and Fernández-Giráldez, 2012). **A:** Linear calcification in the CIMT. **B:** Calcified plaque with posterior acoustic shadowing.

2.9.1.2 Interpretation of IMT results

As the IMT increases in diameter, so does the severity of the disease (Table 2.6) (Hennerici *et al*., 2006). Both the right and left carotid artery IMT should be measured. Within the last ten years, limited studies were published evaluating CIMT parameters. For this reason, the classification described by Hennerici *et al*. (2006) is still used (Table 2.6). However, these parameters correlate with the European Society of Cardiology (Simonova, 2015). As

mentioned previously, IMT measurements can also be geographically influenced (Song *et al.*, 2020). It is, therefore, essential to encourage population-specific studies to determine applicable reference ranges for CIMT.

Table 2.6. Severity classification of IMT measurements (Hennerici *et al*., 2006).

IMT measurement (mm)	Classification
A: <0.9 mm	Normal IMT
B: 0.9–1.1 mm	Mildly thickened IMT
C: 1.2–1.6 mm	Moderately thickened IMT
D: 1.7 mm–1.9 mm	Severely thickened IMT
E: >1.9 mm	Transition from thickened IMT to plaque

The assessment of the number, characteristics, and size of plaques is important. Assessing maximal plaque thickness is important because plaque scores determine the severity of atherosclerosis and CVD (Nezu *et al.*, 2016). Furthermore, the carotid plaque type (homogeneous, heterogeneous, and irregular) is associated with the incidence of ischemic stroke *(Hennerici et al., 2006)*.

According to Costanzo *et al.* (2010), a thickened CIMT predicts the risk of CV events with more substantial predictive power for cerebral vascular events than coronary vascular events. A clinical manifestation of subclinical atherosclerosis is an increased CIMT. For this reason, an increased CIMT has been included in the list of organ damage conditions in the European Hypertension and Prevention Guidelines (Zyriax *et al.*, 2021; Ren *et al.*, 2015).

2.9.2 Radial artery IMT

Significant coronary artery narrowing on diagnostic angiography is associated with an increased radial IMT, and an increased radial IMT is associated with a greater risk of developing future cardiovascular events (Eklund *et al.*, 2013). The radial artery IMT can be measured by using a multi-frequency linear probe. The IMT should be measured about 1-2 cm proximal to the skinfold separating the Palma manus from the regio antebrachial anterior (Figure 2.8) (Myredal *et al.*, 2009). Eklund *et al.* (2013) reported that the radial artery IMT in healthy individuals is <0.3 mm and was also the reference value used in this study.

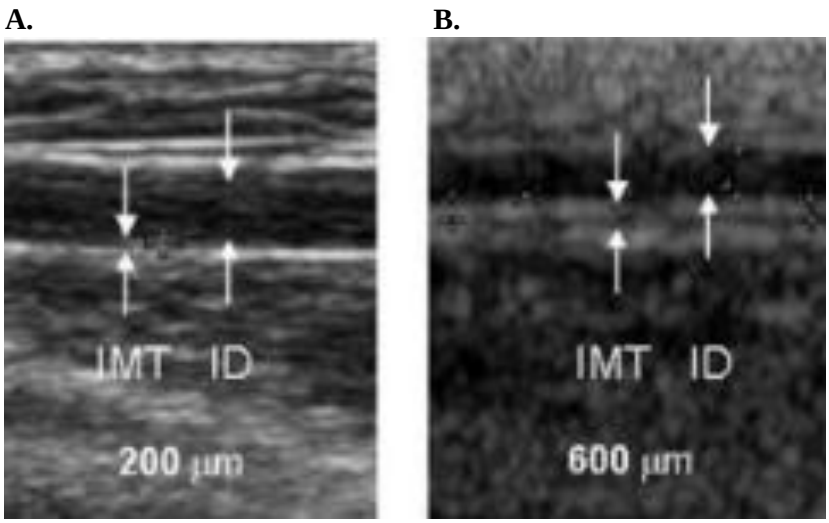
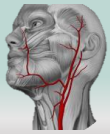


Figure 2.8. *B-mode ultrasonography of the RIMT and internal diameter of the radial artery* (Ku *et al.*, 2006). **A:** Normal RIMT measurement. **B:** Abnormal RIMT measurement.



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Correlation of Cardiac risk factors with carotid intima-media thickness and radial intima-media thickness measurements

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ABSTRACT

Background: Early identification of patients at risk for future cardiovascular events is essential.

Objectives: To evaluate the relationship between cardiovascular risk factors and carotid intima-media thickness (CIMT) and radial intima-media thickness (RIMT).

Methods. A prospective, descriptive-analytical study design was used. Two hundred and fifty patients with one or more modifiable risk factors underwent an ultrasound measurement of left and right CIMT and RIMT according to standard guidelines.

Results. Eighty-one percent of patients had 2 or more risk factors. Hypertension was the most common modifiable risk factor (89%) followed by obesity (66%). Male gender demonstrated a significant increase in mean CIMT ($p< 0,05$). Hypertension, diabetes mellitus (DM), hypercholesterolemia and smoking contributed to a thickened CIMT mean with odds ratios of 3.99, 2.82, 2.47 and 2.09, respectively ($p < 0.05$). Combinations associated with a thicker mean CIMT included hypertension and DM, hypertension and smoking - odds ratios of 6.92 and 3.67 ($p< 0.05$)**.** Only hypercholesterolemia was significantly associated with a thicker mean RIMT ($p<0.05$).

Conclusion. Male sex, increased age, hypertension, DM, hypercholesterolemia and smoking significantly contributed to a thickened CIMT, whereas only hypercholesterolemia was associated with a thickened RIMT. Hypertension had the most significant impact on the CIMT mean thickness. Combinations of risk factors appeared to add summative risks for thickened CIMT and RIMT.

Introduction

Early identification of patients at risk for future cardiovascular events is essential since addressing these early stages is more effective than treating advanced atherosclerotic vascular disease.[1] Smoking, hyperlipidemia, metabolic syndrome, hypertension, and diabetes mellitus (DM) are major cardiac risk factors contributing to intracranial atherosclerosis.[2] The presence of multiple risk factors increases the development of atherosclerosis significantly, which leads to stroke as a result of ruptured plaques.[3]

Raised levels of low-density lipoprotein (LDL) are a dominant cause of atherosclerosis. However, atherosclerosis can also develop due to or in combination with other risk factors such as hypertension, DM, smoking, male sex, and family history. Plaque rupture is multi-layered, involving foam cell formation, smooth muscle cell proliferation, angiogenesis, inflammation, arterial remodelling, thrombosis, fibrous cap rupture, and more. Since plaque formation is heterogeneous, it leads to erratic progression rates and variable outcomes. Although most plaques remain subclinical, some may become obstructive, and others may cause thrombosis, leading to a stroke or acute coronary syndrome (ACS).[4]

Stroke is the second most common cause of mortality and the third most common cause of disability.[5] Eighty percent of strokes are ischemic, while hemorrhagic types are less common, contributing 25-50%.[6] Sacco et al. (1997) emphasized that the most important modifiable risk factor for ischemic stroke is hypertension; hypertension is a relative risk for stroke when the systolic blood pressure ≥160 mmHg and/or diastolic blood pressure ≥95 mmHg.[7]

Patients with DM have a 2-fold increase in stroke risk. Strokes account for approximately 20% of deaths in diabetic patients.[8] The risk of ischemic stroke increases in patients with elevated low-density lipoprotein-cholesterol levels. Patients with low high-density lipoprotein-cholesterol (HDL-C) have a greater stroke risk than healthy individuals.[9] Cigarette smoking is a significant risk factor contributing to almost 15% of all annual stroke deaths.[8] The single most important non-modifiable risk factor for stroke is age, and for each successive ten years after age 55 in both men and women, the stroke rate doubles. More women die of stroke each year because they tend to live longer than men; however, in men, the stroke incidence rates are 1.25 times higher than in women.[7]

Globally, ultrasound has been the most common method of evaluating carotid atherosclerosis due to reasonably low-cost ultrasound analyses, easy access to the carotid artery, and readily available equipment.[9] Carotid intima media thickness (CIMT) is an independent predictor of stroke, although there is limited data on whether an increased IMT predicts stroke. Shimoda et al. (2020) showed that CIMT was positively associated with the risks of stroke, cardiovascular disease, and coronary artery disease (CAD) in the Japanese population.[10] Tsivgoulis et al. (2006) concluded that a higher risk of long-term stroke recurrence is associated with increased CIMT values – the probability of experiencing recurrent strokes increased by 18.0% for each increase of 0.1 mm in CIMT.[11] Carotid ultrasound is one of the few non-invasive imaging techniques that allow the evaluation of vascular function and anatomy.[12] This technique can measure various parameters including vessel diameter, IMT, presence and diameter of stenosis and carotid plaques. Increased thickness of the carotid intima and the presence of large plaques can predict future cardiovascular events.[9]

CIMT is a modest but independent predictor of CAD. Radial intima media thickness (RIMT) is related to several cardiovascular risk factors and can be imaged with excellent precision.[12] Increased occurrence of significant coronary artery narrowing is associated with an increased RIMT. Patients have a nearly three-fold increased risk for major adverse cardiovascular events with RIMT values above the median of 0.3 mm.[13] Therefore, RIMT is a significant predictor of major adverse cardiovascular events, along with type II diabetes and body mass index (BMI).[14] Myredal et al. (2009) reported a 19% elevation in RIMT in patients with coronary heart disease compared to healthy subjects.[14]

Therefore, this study aimed to evaluate which risk factors contribute to increased CIMT and RIMT and whether a correlation exists between risk factors and CIMT and RIMT measurements in patients from central South Africa (SA).

Methods

Study design, location and population

The study design was a prospective, descriptive-analytical single-centre study and was conducted at a private physician practice in Bloemfontein. Patients were referred from the Free State, Northern Cape, and Lesotho. The study sample included 250 first-time visiting patients presenting with one or more modifiable cardiac risk factors from 1 September 2020 to 30 September 2021.

Clinical data

Demographic (age, sex, ethnicity, and geographic location), anthropometric height (cm), weight (kg) and BMI (kg/m²), clinical (modifiable and non-modifiable risk factors and family history and other) and ultrasound data (carotid and radial intimal medial thickness) were recorded for each patient that consented to participate in the study. The modifiable risk factors recorded included obesity, diabetes mellitus, smoking, hypertension, and hypercholesterolemia, and non-modifiable risk factors included age, gender, ethnicity, and family history of CAD. Clinical data recorded from the patient's medical file included HIV status and rheumatoid arthritis (RA).

Definitions

Obesity: Patients were classified as overweight if they had a BMI of ≥25 kg/m² and obese ≥30 kg/m².[15]

Diabetes Mellitus: Type 1 diabetes/Type 2 diabetes as specified in the patient's medical file.

Smoking: Smoking refers to the use of any tobacco product. Both current and previous smokers were regarded as smokers.

Hypertension: Hypertension was defined as a systolic blood pressure ≥140 mmHg and/or diastolic blood pressure ≥90 mmHg.[16]

Hypercholesterolemia: Hypercholesterolemia was defined as total cholesterol ≥6.5 mmol/L.[15]

Clinical investigations

Vascular ultrasound of carotid arteries: Intima-media thickness

All ultrasound investigations was carried out by a single qualified and experienced ultrasonographer. The radial and carotid ultrasound examination was performed according to International Guidelines.[15] Both the right and left carotid IMT were measured, and the mean IMT was calculated and analyzed. All images were obtained in a supine position using a Philips CX50 diagnostic ultrasound system equipped with a 7.5 MHz multi-frequency linear probe. The carotid bulbus were located, and the CIMT was measured 10 mm proximal to the bulbus. CIMT measurements were classified as normal (<0.9mm) and abnormal (≥0.9 mm).[15]

Vascular ultrasound of the radial artery

The right and left radial artery IMT was measured about 1-2cm proximal to the skinfold separating the palma manus from the regio antebrachia anterior.[14] The mean RIMT was calculated and analyzed. A radial IMT >0.3mm was considered abnormal.[15]

Ethical Clearance

Ethical clearance was obtained from the Health Sciences Research and Ethics Committee (HSREC) of the University of the Free State (**UFS-HSD2020/2018/2601)**. Standard informed consent was obtained in all subjects.

Statistical analysis

All recorded data were captured in a datasheet using Microsoft® Excel® 2019. Data were quality controlled by an independent investigator for accuracy and transcription errors. The statistical analyses were performed using the R program. Continuous data were summarised using descriptive statistics, including but not limited to mean, standard deviation (SD), median, minimum, maximum and interquartile range (IQR). Descriptive analyses were carried out per subgroup depending on the number of patients. Linear regression was used to test for the effect of the covariates on the response variables, and logistic regression was used to determine the relative risk. Categorical data were summarized utilizing frequency tables. The cell counts were compared using a Chi^2 test or Fisher's exact test. The latter was used if one or more of the cell counts were less than 5. A p-value of $< 0,05$ was considered as statistically significant. Correlations were obtained using the Pearson correlation coefficient test.

Results

Two hundred and fifty patients met the inclusion criteria. As expected, most patients came from the Free State (97.2%) and only 2.8% from other provinces. Most of these patients resided in the Mangaung Metropolitan district (85.6%). The mean age of patients was 60.2 ± 16.5 years, with a female predominance (63.6% vs 36.4%), and most patients were Caucasian (76%).

In the total study group, 42% (n=104) of patients presented with three or more modifiable risk factors, 39% (n=98) with two risk factors, and 19% (n=48) with only one modifiable risk factor. Hypertension was the most common modifiable risk factor (89%), followed by obesity (66%). DM was present in 30% of patients, of which 55% presented with DM type 1 and 45% with DM type 2.

Carotid-intima media thickness

The mean CIMT of the total study group was 0.8±0.2 mm, and the maximum CIMT was 1.9 mm (right carotid artery) and 1.7 mm (left carotid artery). The mean CIMT in men was 0.9±0.2 mm and in women 0.8±0.3 mm. A transient ischemic attack (TIA) or stroke was recorded in 24% (n=61) of the study population, and 51% (n=55) of patients diagnosed with a stroke or TIA had an abnormal CIMT (≥ 0.9 mm).

An abnormally thickened CIMT (≥0.9 mm) was observed in 107 (43%) patients with a mean CIMT of 1.1±0.15 mm. The demographic, anthropometric, and modifiable risk factors of patients with normal and abnormal CIMT are summarized in Table 1.

Non-modifiable risk factors

The mean age of patients with an abnormal CIMT was similar to patients with a normal CIMT (62.4±16.4 years vs 60.1±16.5 years, respectively, p=0.23). Significantly more men presented with an abnormal CIMT compared to the normal CIMT group (p<0.05). The number of Caucasians in both groups was similar. A positive family history of cardiovascular disease was recorded in 89% of patients with an abnormal CIMT (Table 1).

Modifiable risk factors

Almost two-thirds of patients (63%, n=67) with abnormal CIMT measurements had three or more modifiable risk factors, 31% (n=33) with two risk factors, and only 6% (n=7) presented with one modifiable risk factor.

An abnormal CIMT was recorded in 50% (n=112) of hypertensive patients. Hypertension was the leading modifiable risk factor and was present in almost all patients with an abnormal CIMT (95%). Hypertension was followed by obesity (66%), hypercholesterolemia (51%), DM (44%), and smoking in 28% of patients. The BMI did not significantly differ between the abnormal and normal CIMT groups (28.9±6.9 kg/m^2 vs 27.9±6.8 kg/m^2; p=0.99) (Table 1).

Table 1. Demographic, anthropometric and modifiable risk factors of patients with an abnormal CIMT

Variable (unit)	Hypertension	DM	Hyper-cholesterolemia	Obesity	Smoking
Normal CIMT (n=143)	121	26	37	89	29
Age (y; mean±SD)	60.4±16.3	60.8±16	60.3±16.3	60.3±16.3	60.1±16.5
Sex:					
Male	35(28.9%)	7(27%)	8(21.6%)	28(31.5%)	12(41.3%)
Female	86(71.1%)	19(73%)	29(78.4%)	61(68.5%)	17(58.7%)
Ethnicity (n;%):					
Caucasian	85(70%)	15(57.7%)	25(67.6%)	62(70%)	27(93.1%)
Mixed Race	4(3%)	0	1(2.7%)	2(2.2%)	1(3.4%)
Black African	32(27%)	10(38.5%)	11(29.7%)	24(27%)	1(3.4%)
Asian	0	1(3.8%)	0	1(2.2%)	0
Indian	0	0	0	0	0
BMI (kg/m^2; mean±SD):	29.0±6.9	29±6.9	29±7.0	29 ± 7.0	28.9±6.9
Normal	44(36.4%)	6(23%)	8(21.6%)		13(44.8%)
Overweight	77(63.6%)	20(77%)	29(78.4%)		16(55.2%)
Abnormal CIMT (n=107)	102	48	55	71	30
Age (y; mean±SD)	60.4 ± 16.4	61 ± 6.0	60.4 ± 16.4	60.4 ± 16.4	61.9 ± 16.4
Sex:					
Male	46(45%)	27(56.2%)	29(52.7%)	34(47.9%)	20(66.7%)
Female	56(55%)	21(43.8%)	26(47.3%)	37(52.1%)	10(33.3%
Ethnicity (n;%):					
Caucasian	82(80.1%)	36(75%)	47(85.5%)	57(80.3%)	25(83.3%)
Mixed Race	4(3.9%)	2(4.2%)	3(5.5%)	4(5.6%)	1(3.3%)
Black African	15(14.7%)	10(20.8%)	4(7.3%)	10 (14.1%)	3(10%)
Asian	1(0.9%)	0	0	0	0
Indian	0	0	1(1.8%)	0	1(3.3%)
BMI (kg/m^2; mean±SD):	28.9 ± 6.9	29 ± 6.7	28.9 ± 6.9	28.9 ± 6.9	29.0 ± 6.9
Normal	33(32.4%)	16(33.3%)	23(41.8%)		12(40%)
Overweight	69(67.6%)	32(66.7%)	32(58.2%)		18(60%)

[CIMT, carotid intima media thickness; y, years; SD, standard deviation; BMI, body mass index; kg, kilogram; DM, diabetes mellitus]

The non-modifiable risk factors analysis showed that more men with hypertension, DM and hypercholesterolemia had abnormal CIMT measurements. In contrast, females dominated the normal group (Fig. 1).

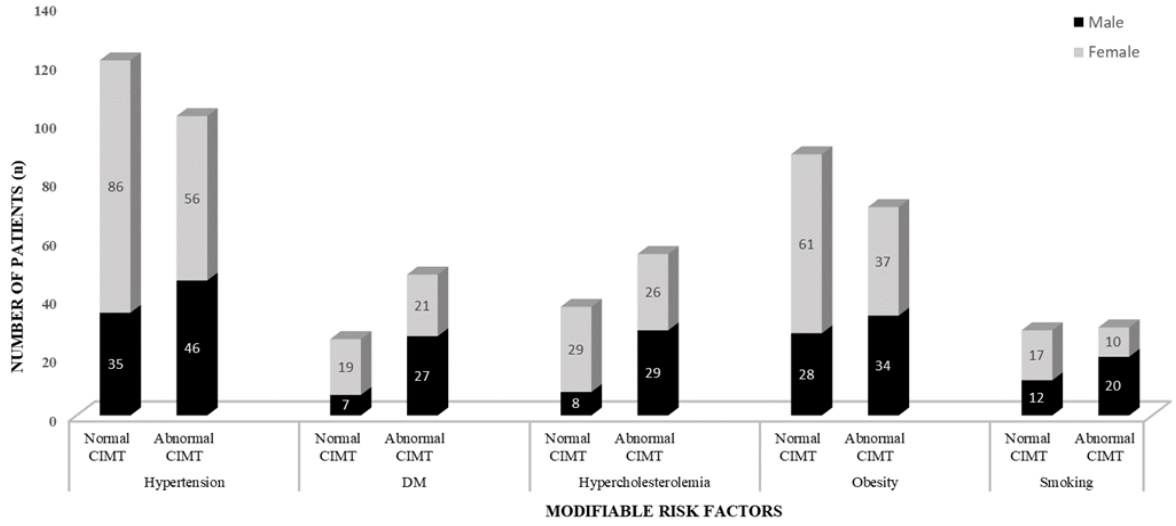

Fig. 1. Distribution of gender for normal and abnormal CIMT in modifiable risk factor groups.

Effect of risk factors on CIMT

The mean CIMT in patients with DM, hypertension, hypercholesterolemia, and smoking was significantly increased compared to those without. (Table 2). Male gender was associated with a significant increase in mean CIMT ($p < 0,05$). Age had some effect, and analyses projected that each year of ageing added 11% to the risk of having a thickened mean CIMT. This is supported by the fact that only 4 people under 50 years presented with an abnormal CIMT. The other non-modifiable risk factors did not contribute to a mean increase in CIMT. The odds ratios of modifiable risk factors associated with an increase in mean CIMT are represented in Table 2.

Table 2. The effects of individual modifiable risk factors on CIMT and risk associated with increased CIMT

Modifiable risk factor	Mean CIMT (mm)	95% CI	OR	95% CI
Hypertension	0.76*	0.05, 0.21	3.99	1.50, 12.7
Diabetes Mellitus	0.74*	0.06, 0.17	2.82	1.53, 5.24
Hypercholesterolemia	0.73*	0.05, 0.15	2.47	1.39, 4.40
Smoker	0.72*	0.03, 0.14	2.09	1.07, 4.17
Obesity	0.64	-0.04, 0.06	1.06	0.60, 1.88

*** Indicate statistical significance = .** [CIMT, carotid intima-media thickness; mm, millimetre; CI, confidence interval; OR, odds ratio;]

Several modifiable risk factor combinations were evaluated to assess whether an association with a thicker mean CIMT was present. The only risk factor combinations that statistically contributed to a thicker CIMT mean were DM and hypertension; DM and hypercholesterolemia; hypertension and smoking; hypercholesterolemia and obesity (Table 3).

Unexpectedly, the combination of hypercholesterolemia and DM and hypercholesterolemia and obesity appeared to reduce the risk of an increased mean CIMT and might have a protective effect. More than 50% of patients diagnosed with hypercholesterolemia were on anti-hypercholesterol treatment (Table 3).

Table 3. The effects of modifiable risk factor combinations on CIMT mean

Modifiable risk factor combinations	linear regression		logistic regression	
	p-value	95% CI	OR	95% CI
Diabetes mellitus + hypertension*	0.018	0.07, 0.75	6.92	2.32, 20.7
Hypertension + smoker*	0.033	0.02, 0.52	3.67	1.43, 9.41
Diabetes mellitus + hypercholesterolemia*	0.019	-0.24, -0.02	0.15	0.03, 0.70
Hypercholesterolemia + obesity*	0.003	-0.27, -0.06	0.12	0.03, 0.51

*Only the results of statistical significance are reported in the table above. [CIMT, carotid intima-media thickness; mm, millimetre; CI, confidence interval; OR, odds ratio]

Radial intima-media thickness

A minority of the study population (22%, n=55/250) had an abnormal thickened RIMT. As expected, the mean internal radial diameter of the abnormal RIMT group (2.6±0.5 mm) was significantly thicker than the normal RIMT group (2.3±0.5 mm $p<0.05$). Only 38% of patients with abnormal CIMT also had an abnormal RIMT.

The mean age of the abnormal and normal RIMT patient groups was similar (61±16.1 vs 60±16.5 years, respectively). In contrast to CIMT, more female patients presented with an abnormal RIMT than men (57% vs 43%), and most patients were Caucasian (82%). Most patients in the abnormal RIMT group had a positive family history of ischemic heart disease (82%).

Similar to patients with abnormal CIMTs, hypertension was the most common modifiable risk factor in the abnormal RIMT group (91%), followed by obesity (73%) and hypercholesterolemia (56%). DM was present in 36% of patients with an abnormal RIMT. Hypercholesterolemia was the only individual risk factor that significantly contributed to a thicker mean radial IMT ($p<0.05$). The only risk factor combinations that significantly contributed to a thicker mean RIMT were hypertension and obesity, DM and obesity and hypertension and hypercholesterolemia (Table 4).

Table 4. The effects of modifiable risk factors and combinations on RIMT mean

Modifiable risk factor combinations	linear regression		Logistic regression	
	p-value	95% CI	OR	95% CI
Hypertension*	0.5	0.00, 0.09		0.00, 0.09
Hypercholesterolemia*	<0.001	0.02, 0.08		0.02, 0.08
Hypertension + obesity*		0.05, 0.15	0.13	0.03, 0.61
Hypertension + hypercholesterolemia*	0.004	0.03, 0.14	2.87	1.41, 5.84
Diabetes mellitus + obesity*	0.009	-0.04, 0.06	3.64	0.69, 19.2

*Only the results of statistical significance are reported in the table above. [RIMT, radial intima-media thickness; mm, millimetre; CI, confidence interval; OR, odds ratio]

Correlations

A strong correlation was observed between carotid IMT left and right and radial IMT left and right. However, there was a poor correlation between CIMT and RIMT (Table 5). The latter is not unexpected, as only 38% of patients with abnormal CIMT had an abnormal RIMT.

Table 5. Correlation between CIMT and RIMT left and right

	Carotid IMT left*	Radial IMT right*	Radial IMT left*
Carotid IMT right	0.93	0.47	0.48
Carotid IMT left	-	0.42	0.43
Radial IMT right	-	-	0.98

* correlation co-efficient(r) for respective IMT's

Interestingly, 40 patients in the study presented with rheumatoid arthritis. In this subgroup, 70% of patients had an abnormal CIMT. The numbers were too small to analyze further, and the study was not designed to assess co-morbidity risks.

Discussion

The prognostic and predictive value of CIMT measurements and cardiovascular disease is a topic that is hotly debated in current literature. [17] It is, however, clear that an increase in IMT thickness is an independent predictor for the risk of stroke apart from the traditional well-described risk factors [15]. Only patients with individual risk factors for future cardiovascular system (CVS) events, such as stroke and CAD, were included in this study. This is the first study describing CIMT and RIMT ultrasound measurements in patients with modifiable risk factors in central SA.

The study group primarily consisted of older, affluent patients presenting at a general physician private practice which explains the demographic composition. The CIMT diameters of the total study group, men and women, were similar to the findings of a study conducted by Mostaza *et*

al. (2018).[16] Despite the presence of several risk factors, 80% of patients had two or more risk factors. Only 40% of patients with risk factors had pathological thickening of the carotid intima. However, most patients with a CIMT of ≥0.9 mm had two or more risk factors. It is perturbing that hypertension was present in more than 90% of patients, and hypertension was the single most common risk factor observed in the study population. Two-thirds were obese, followed by hypercholesterolemia, diabetes and smoking. Male sex was the only non-modifiable risk factor associated with abnormal CIMT measurements. As a matter of fact, where females tended to be dominant in the normal CIMT group, an almost 50/50 distribution of gender was observed for those with an abnormal CIMT. More than 50% of patients diagnosed with stroke or transient ischemic attack (TIA) had an abnormal CIMT. This finding concurs with Roquer *et al*. (2011), who demonstrated that an increase in IMT thickness is an independent marker of major cardiovascular events. For each increment of 0.1 mm in IMT, the probability of experiencing a recurrent stroke is increased by 18%.[17]

Our results show that individual risk factors played a role in a thickened CIMT. However, the high prevalence of hypertension in central South Africa is cause for concern. Hypertension was markedly higher in this study (90%) than that reported by Ren et al. and Dias *et al*. (67% and 72%, respectively) [3,18]. Forty-six percent of the hypertension group presented with an abnormal CIMT. Interestingly, only 20% of the hypertension group in the study of Ren et al. (2015) presented with carotid atherosclerosis.[3] In our study, hypertension contributed to a thicker CIMT mean with an OR of 3.99. This result corresponds to the findings of Ferreira et al. (2016), demonstrating that individuals with a CIMT ≥0.9 mm had a significant association with a hypertension history, with an OR of 2.11.[19] Shear forces produced by chronically high blood pressure damage the endothelial layer in cerebral arteries, coronary arteries and the aorta.[20] According to Shrestha *et al*. (2009), an increase in systolic blood pressure correlates with atherosclerotic changes in the carotid arteries. An association with low and high-grade lesions correlates with increased systolic blood pressure.[21] Patients with cognitive dysfunction that are hypertensive have changes in the vascular morphology supported by an increased carotid IMT, enhanced atherosclerotic lipid profile and impaired hemodynamic function. This is manifested by elevated central systolic blood pressure. Increased systolic blood pressure causes morphological changes characterized by an increased carotid IMT.[22]

Diabetes was a prominent risk factor and was recorded in 30% of the study population. The prevalence of diabetes in our study is lower than the findings of Poznyak *et al*. (2013), who

investigated 3001 patients in South Africa and reported a 49.5% prevalence of DM.[22] In 60% of our patients with DM, an abnormal CIMT was recorded. The mean CIMT in the diabetic patients was 0.9±0.2 mm concurring with Yafei *et al*. (2019).[23] They reported that CIMT were significantly thicker in diabetic patients compared to controls (0.79±0.16 mm vs 0.58±0.08 mm).[24] In our study, DM significantly contributed to a thicker CIMT mean with an OR of 2.82. This agrees with Ren *et al*. (2015), who showed that diabetes is one of the significant risk factors associated with carotid atherosclerosis.[3] All types of diabetes have been shown to be independent risk factors for accelerated atherosclerosis development.[24] The pathogenesis of DM and atherosclerosis are closely linked, and among the known pathological mechanisms linking atherosclerosis and DM are hyperglycemia, inflammation, increased oxidative stress and dyslipidemia.[24]

Hypercholesterolemia was present in more than half of the patients in the abnormal CIMT group. Interestingly, 86% of patients that presented with hypercholesterolemia and an abnormal CIMT were Caucasian, and only 7% were black Africans. Hypercholesterolemia contributed to a thickened CIMT with an OR of 2.47; this finding has also been reported in several other studies.[3] [24] [25]

Our study showed that risk factor combinations like DM and hypertension, and hypertension and smoking significantly increase the risk of having thicker CIMT measurements. The odds ratios of the combinations were markedly higher than isolated risk factors, most likely indicating the summative effects when individual risk factors occur in combination. However, it appears that having hypercholesterolemia and DM or hypercholesterolemia and obesity reduces the risk of having a thickened CIMT. The authors speculate that many of these patients were on statin treatment which may have provided protection against an increased CIMT. According to Lind (2020), statin treatment is known to reduce cardiovascular events and age-related progression of IMT of the carotid artery in subjects with and without cardiovascular disease.[24] A real-life observational study showed that statin treatment reduced the increase in IMT seen over 10 years compared to subjects not treated with statins. This is further supported by Kerola *et al*. (2021), who concluded that in hypercholesterolemic adults with subclinical atherosclerosis, one-year treatment with rosuvastatin significantly reduced the CIMTs bilaterally and improved the lipoprotein and lipid levels.[25]

Age and male sex were the only non-modifiable risk factors contributing to the risk of an increase in mean CIMT: each year of age added 11% to the risk of having a thickened CIMT. This finding concurs with Ren *et al*. (2015); in this study, the authors demonstrated that middle-aged and older individuals with cardiac risk factors displayed an increased CIMT and higher severity grade than the younger age groups. [3] Interestingly, Zyriax *et al*. (2021) found that each year of life translated to an additional 0.004 mm increase in CIMT. Apart from demonstrating a risk for thicker CIMTs, significantly more men had an abnormal CIMT compared to the normal CIMT group indicating male sex is a noteworthy risk factor.[1] Similar findings were reported by Mostaza *et al*. (2018), where the male gender was associated with a thickened CIMT.[16]

According to, Wahood *et al*. (2022), patients with hypertension and congestive heart failure usually have an increased radial artery diameter.[26] Furthermore, Loh *et al*. (2007) concluded that hypertension, hyperlipidemia and male sex increased the radial artery diameter, whereas age, DM, race and smoking did not significantly influence the size of the radial artery. Vessel dilation is promoted by the release of endothelium-derived nitric oxide (NO) and defective synthesis.[27]

Interestingly, more female patients presented with an abnormal RIMT than men (57% vs 43%). Similar to the CIMT group, hypertension was the most common modifiable risk factor, followed by obesity and hypercholesterolemia. Hypercholesterolemia was the only individual risk factor significantly contributing to a thicker radial IMT with an OR of 3.00. In contrast to CIMT, hypertension and obesity was the only combination contributing to a thicker RIMT. Eklund *et al*. (2013) concluded that RIMT assessed by ultrasound confers prognostic information in patients with suspected CAD. Similar findings were reported by Eklund *et al*. (2012), where RIMT may constitute a feasible imaging biomarker for systemic atherosclerotic burden.[13] Our results demonstrate a poor correlation between RIMT and CIMT. The authors speculate that RIMT may be more valuable as a potential marker of coronary atherosclerotic disease while CIMT appears to be more valuable for neurovascular disease – this will provide some explanation for the difference in association with risk factors found in the study.

It is thought-provoking that in this study, forty patients with risk factors also had rheumatoid arthritis as a co-morbidity, and 70% had an abnormal thickened CIMT. According to Hannawi *et al*. (2007), CIMT in RA patients is increased compared to healthy individuals matched for

age, sex and cardiovascular risk.[28] This statement is supported by Komici et al. (2021), showing that CIMT has previously been found to be increased in patients with longstanding RA.[29] Patients with longstanding RA, >20 years, had a higher CIMT compared to patients of similar age but shorter disease duration, <7 years. An important factor related to cardiovascular risk in RA patients is an impairment of endothelial functions, which is a key element in the development of the atherosclerosis process. Endothelial impairment is related to RA being a chronic inflammatory process. Altered endothelial reactivity has been documented in RA patients prior to atherosclerotic plaque detection, even without cardiovascular risk factors.[28]

Study limitations

This study was conducted during the Covid-19 pandemic resulting in relatively small numbers. Patients were recruited from private practice, thus consisting mainly of an affluent population. The lack of long-term follow-up of patients with abnormal CIMT was also regarded as a limitation. Based on the results obtained in this study, further studies are indicated.

Conclusion

Most patients had two or more risk factors, and hypertension was present in almost 90% of patients. Results showed that male sex, increasing age, hypertension, DM, hypercholesterolemia and smoking significantly contributed to a thickened CIMT. In contrast, only hypercholesterolemia was associated with a thickened RIMT. Hypertension had the biggest impact on increased mean CIMT compared to the other modifiable risk factors. Certain risk factor combinations were also associated with a thickened CIMT and RIMT, and combinations of risk factors appeared to add summative risk. A good correlation was observed between left and right CIMT and between left and right RIMT measurements. However, CIMT and RIMT did not correlate with each other.

Conflict of interest

The authors declare that there are no conflicts of interest relating to this study.

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Abbreviations

BMI	Body mass index
CAD	Coronary artery disease
CIMT	Carotid intimal media thickness
CI	Confidence interval
CM	Centimeter
CVS	Cardiovascular system
DM	Diabetes Mellitus
ETC	Et cetera
HDL	High-density lipoprotein
HDL-C	High-density lipoprotein-cholesterol
HSREC	Health Sciences Research and Ethics Committee
HIV	Human immunodeficiency virus
IMT	Intima media thickness
Kg	Kilogram
Kg/m^2	Kilogram-Meter Squared
LDL	Low-density lipoprotein
LDL-C	Low-density lipoprotein-cholesterol
MHz	Megahertz
mm	Millimeter
mmHg	Millimeter of mercury
mmol/L	Millimoles per litre
NO	Nitric Oxide
OR	Odds ratio
RA	Rheumatoid arthritis
RIMT	Radial intimal media thickness
SA	South Africa
SD	Standard deviation

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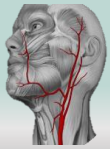
Chapter 4 – General Conclusion



In this study, we aimed to investigate the possible relationship between CIMT and RIMT and modifiable and non-modifiable cardiac risk factors. Only patients with risk factors for future CVS events, such as CAD and stroke, were included in the study. To our knowledge, this is the first study describing CIMT and RIMT ultrasound measurements in patients with modifiable risk factors in central SA. To date, there is a lot of debate in the literature regarding the predictive and prognostic value of CIMT measurements. However, it is clear that apart from traditional cardiac risk factors, an increase in CIMT thickness is an independent predictor of stroke risk.

The study group primarily consisted of older, affluent patients who presented at a private practice with one or more modifiable cardiac risk factors. Male sex and increased age significantly contributed to a thickened CIMT. Only 40% of patients with risk factors had a thickened CIMT. The majority of patients in the abnormal CIMT group had two or more risk factors. Hypertension was the single most common risk factor in the study population, present in 89%; two-thirds were obese, followed by hypercholesterolemia, diabetes and smoking. Females tended to be dominant in the normal CIMT group, but almost a 50/50 distribution of gender was observed for the abnormal CIMT group. Nearly 60% of patients diagnosed with stroke or TIA had an abnormal CIMT.

Forty-six percent of the hypertension group had an abnormal CIMT (>0.8 mm), and hypertension contributed to a thicker CIMT mean (OR 3.99). Diabetes was an important risk factor in our study population, and 60% of our patients presenting with DM had an abnormal CIMT. The mean CIMT in the diabetic patients was 0.9±0.2 mm. DM significantly contributed to a thicker CIMT mean (OR 2.82). Hypercholesterolemia was present in more than half of the patients in the abnormal CIMT group. Hypercholesterolemia contributed to a thickened CIMT (OR of 2.47).

Results of this study showed that risk factor combinations like DM and hypertension (OR 6.92) and hypertension and smoking (OR: 3.67) significantly increase the risk of having thicker

CIMT measurements – the odds ratios of the combinations were markedly higher than single risk factors, most likely indicating summative effects when individual risk factors occur in combination. However, it appears that having hypercholesterolemia and DM, hypercholesterolemia and obesity reduces the risk of having a thickened CIMT. The authors speculate that many of these patients were already on statin treatment which may have provided protection against an increased CIMT.

A good correlation was observed between left and right CIMT and between left and right RIMT measurements. However, CIMT and RIMT did not correlate with each other. Abnormal RIMT measurements were observed in only 22% of the study population, and only 38% of patients with abnormal CIMT also had an abnormal RIMT, indicating that these are not necessarily correlated. Interestingly, the mean internal radial diameter was significantly smaller in the normal RIMT group compared to the abnormal RIMT group (2.3±0.5 mm vs 2.6±0.5 mm, respectively). It is important to take cognizance of the radial artery diameters described in this study because radial artery catheterizations are increasingly being performed in these patients due to low complication rates and significantly reduced procedure times.

Interestingly, more female patients presented with an abnormal RIMT than men (57% vs 43%). Like the CIMT group, hypertension was the most common modifiable risk factor, followed by obesity and hypercholesterolemia. Hypercholesterolemia was the only individual risk factor contributing to a thicker radial IMT (OR 3.00), and, in contrast to CIMT, hypertension and obesity was the only combination that contributed to a thicker RIMT. Forty patients with risk factors also had Rheumatoid Arthritis as a co-morbidity, and 70% had an abnormal thickened CIMT. A high percentage of patients in the RA group presented with an abnormal CIMT, which warrants further investigations.

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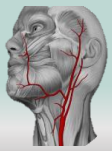

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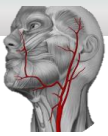
Appendices



Appendix A

UNIVERSITY OF THE FREE STATE
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YUNIVESITHI YA FREISTATA

UFS·UV
HEALTH SCIENCES
GESONDHEIDSWETENSKAPPE

Health Sciences Research Ethics Committee

19-Jan-2021

Dear **Mr Fritz Van Schalkwyk**

Ethics Clearance: **Carotid and radial intima-media thickness measurements in private patients presenting with modifiable cardiac risk factors in Central South Africa**

Principal Investigator: **Mr Fritz Van Schalkwyk**

Department: **Clinical Technology - CUT**

Submission Page

APPLICATION APPROVED

Please ensure that you read the whole document

With reference to your application for ethical clearance with the Faculty of Health Sciences, I am pleased to inform you on behalf of the Health Sciences Research Ethics Committee that you have been granted ethical clearance for your project.

Your ethical clearance number, to be used in all correspondence is:**UFS-HSD2020/2018/2601**

The ethical clearance number is valid for research conducted for one year from issuance. Should you require more time to complete this research, please apply for an extension.

We request that any changes that may take place during the course of your research project be submitted to the HSREC for approval to ensure we are kept up to date with your progress and any ethical implications that may arise. This includes any serious adverse events and/or termination of the study.

A progress report should be submitted within one year of approval, and annually for long term studies. A final report should be submitted at the completion of the study.

The HSREC functions in compliance with, but not limited to, the following documents and guidelines: The SA National Health Act. No. 61 of 2003; Ethics in Health Research: Principles, Structures and Processes (2015); SA GCP(2006); Declaration of Helsinki; The Belmont Report; The US Office of Human Research Protections 45 CFR 461 (for non-exempt research with human participants conducted or supported by the US Department of Health and Human Services- (HHS), 21 CFR 50, 21 CFR 56; CIOMS; ICH-GCP-E6 Sections 1-4; The International Conference on Harmonization and Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH Tripartite), Guidelines of the SA Medicines Control Council as well as Laws and Regulations with regard to the Control of Medicines, Constitution of the HSREC of the Faculty of Health Sciences.

For any questions or concerns, please feel free to contact HSREC Administration: 051-4017794/5 or email EthicsFHS@ufs.ac.za.

Thank you for submitting this proposal for ethical clearance and we wish you every success with your research.

Yours Sincerely

Prof. A. Sherriff
Chair : Health Sciences Research Ethics Committee

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