Sudan University of Science and Technology
College of Graduate Studies

Assessment of Serum High Sensitive C-Reactive Protein Level among Patients with Metabolic Syndrome – Khartoum State

تقييم مستوى البروتين C التفاعلي عالي الحساسية في مصل الدم لدى مرضى متلازمة التمثيل الغذائي – ولاية الخرطوم

A dissertation submitted in partial fulfilment for the requirement of M.Sc. degree in Medical Laboratory Sciences (Clinical Chemistry)

By:

Mohammed Eltayeb Omer Ibrahim Mohammed

B.Sc. in Medical Laboratory Sciences-Clinical Chemistry

(Omdurman Islamic University 2017)

Supervisor:

Dr. Mariam Abbas Ibrahim

Sudan University of Sciences and Technology

College of Medical Laboratory Sciences

Clinical Chemistry Department

December-2020

بِسمِ اللهِ الرَحمنِ الرَحِيمِ

قَالَ تَعَالَى :

﴿وَ إِذَا مَرِضْتُ فَهُوَ يَشْفِينِ﴾

سورة الشعراء،الآيـة : ﴿٨٠﴾

Dedication

To symbol of love & giving my parents

……..

To every patient suffers from MetS

……

To the origin of creation and excellence SUST

Acknowledgments

First, I thank Allah who helps me to accomplish this work.

I would like to express my appreciation to my supervisor Dr. Mariam Abbas Ibrahim who has cheerfully answered my queries, provided me with materials, checked my examples, assisted me in a myriad ways with the writing and helpfully commented earlier draft of this project. I am very proud to join you in this project; it's really great honour for me.

My gratitude extended to the staff of Alriada medical laboratory for their cooperation, great help and technical support through this study.

Abstract

Worldwide prevalence of metabolic syndrome (MetS) is widely diverse, ranges from <10% to 84%. MetS is associated with increased Cardiovascular disease (CVD) mortality. Elevated levels of high-sensitivite C-reactive protein (Hs-CRP) are associated with increased risk for CVD, this study aimed to assess serum level of High-sensitive C-reactive protein in patients with MetS.

This case control study was carried out in Abdalla Khalil center and Mahdi center, in Khartoum state, during the period from April 2019 to November 2020. A total of 66 participants were enrolled in this study, 33 of them were MetS patients as case (16 male and 17 female) and 33 healthy individuals as control (16 male and 17 female), age was matched in both groups. Serum Hs-CRP levels were estimated by using FIA meter. Data were analyzed using SPSS computer program.

The level of Hs-CRP was significantly increased in MetS patients (20.7 ± 14.8) compared to control group (6.38±1.38) with *P*-value 0.000. According to age Hs-CRP level was insignificantly differ in patients who were < 50 years when compared to those who were > 50 years (22.75 ± 15.47 and 18.78 ± 14.34) respectively, with *P*-value 0.452. According to gender Hs-CRP level was insignificantly differ in females (23.76 ± 14.56) compared to males (17.46 ± 14.8) with *P*-value 0.227. According to BMI Hs-CRP level was insignificantly differ in obese patients (≥ 30 Kg/m^2) compared to overweight patients (25-29.9 Kg/m^2) (21.1 ± 12.1 and 20.4 ± 17.04) respectively, with *P*-value 0.162. According to therapy usage Hs-CRP level was significantly decreased in patients who were using lipid-lowering drugs compared to patients who were not (13.5 ± 8.6 and 24.8 ± 16.2) respectively, with *P*-value (0.014), but it was insignificantly differ in patients who were using glucose-lowering drugs

or not (17.7 ± 12.9 vs. 24.2 ± 16.6), and those who were using blood pressure-lowering drug drugs or not (23.1 ± 17.1 vs. 18.1 ± 11.9), with *P*-value (0.216 and 0.351) respectively. There was no correlation between the level of Hs-CRP and both of BMI and WC with (r = 0.079 and -0.118) respectively, but there was +ve, weak and significant correlation between BMI and WC with (r = 0.475).

MetS patients had increased Hs-CRP levels which might be associated with increased risk of CVD.

ملخص الأطروحة

يتنوع انتشار متلازمة التمثيل الغذائي في جميع أنحاء العالم على نطاق واسع، ويتراوح من أقل من 10٪ إلى 84٪، وترتبط متلازمة التمثيل الغذائي بزيادة معدل وفيات أمراض القلب والأوعية الدموية، كما ترتبط المستويات المرتفعة من البروتين سي التفاعلي عالي الحساسية بزيادة خطر الإصابة بأمراض القلب والأوعية الدموية، وتهدف هذه الدراسة إلى تقييم مستوى البروتين سي التفاعلي عالي الحساسية في المرضى الذين يعانون من متلازمة التمثيل الغذائي.

أجريت هذه الدراسة في مركز عبد الله خليل ومركز المهدي بولاية الخرطوم خلال الفترة من أبريل 2019 إلى ديسمبر 2020. وقد تم تسجيل 66 مشاركًا في هذه الدراسة، 33 منهم من مرضى متلازمة التمثيل الغذائي كحالات (16 ذكر و 17 أنثى) و 33 فردًا سليمًا كمجموعة تحكم (16 ذكر و 17 أنثى)، تم مطابقة العمر في كلا المجموعتين، تم تقدير مستويات البروتين سي التفاعلي عالي الحساسية باستخدام مقياس FIA. كما تم تحليل البيانات باستخدام برنامج الكمبيوتر SPSS.

تمت زيادة مستوى البروتين سي التفاعلي عالي الحساسية بشكل ملحوظ في مرضى متلازمة التمثيل الغذائي (20.7 ± 14.8) مقارنة بمجموعة التحكم (6.38 ± 1.38) بقيمة P 0.000. وفقًا للعمر ، كان مستوى البروتين سي التفاعلي عالي الحساسية مختلفًا بشكل طفيف في المرضى الذين تقل أعمارهم عن 50 عامًا بالمقارنة مع أولئك الذين تزيد أعمارهم عن 50 عامًا (22.75 ± 15.47 و 18.78 ± 14.34) على التوالي ، بقيمة P 0.452. وفقًا للجنس ، كان مستوى البروتين سي التفاعلي عالي الحساسية مختلفًا بشكل ضئيل في الإناث (23.76 ± 14.56) مقارنة بالذكور (17.46 ± 14.8) بقيمة P 0.227. وفقًا لمؤشر كتلة الجسم ، كان مستوى البروتين سي التفاعلي عالي الحساسية مختلفًا بشكل طفيف في مرضى السمنة (≤ 30 كجم / م 2) مقارنة بالمرضى الذين يعانون من زيادة الوزن (25-29.9 كجم / م 2) **(21.1 ± 12.1 و 20.4 ± 17.04)**على التوالي ، بقيمة P 0.162. وفقًا لاستخدام العلاج ، انخفض مستوى البروتين سي التفاعلي عالي الحساسية بشكل كبير في المرضى الذين كانوا يستخدمون الأدوية الخافضة للدهون مقارنة بالمرضى الذين لم يكونوا يستخدمونها (13.5 ± 8.6 و 24.8 ± 16.2) على التوالي ، مع قيمة P (0.014) ، ولكن كان هناك اختلاف بسيط في المرضى الذين كانوا يستخدمون عقاقير خافضة

للجلكوز أم لا (17.7 ± 12.9 مقابل 24.2 ± 16.6) ، وأولئك الذين كانوا يستخدمون أدوية خافضة لارتفاع ضغط الدم أم لا (23.1 ± 17.1 مقابل 18.1 ± 11.9) ، بقيمة P 0.216 و0.351على التوالي. لم يكن هناك ارتباط بين مستوى البروتين سي التفاعلي عالي الحساسية وكلا من مؤشر كتلة الجسم ومحيط الخصر مع (r = 0.079 و -0.118) على التوالي ، ولكن كان هناك ارتباط إيجابي ضعيف ومعنوي بين مؤشر كتلة الجسم ومحيط الخصر مع (r = 0.475).

خلصت الدراسة إلى أن مرضى متلازمة التمثيل الغذائي لديهم زيادة في مستويات البروتين سي التفاعلي عالي الحساسية التي قد تترافق مع زيادة خطر الإصابة بأمراض القلب والأوعية الدموية.

List of contents

List of tables

List of figures

List of abbreviations

AHA-NHLBI = American Heart Association/National Heart Lung and Blood Institute

ASCVD = Atherosclerotic cardiovascular disease

AT II = Angiotensin II

BMI = Body mass index

BP = Blood pressure

CHD = Coronary heart disease

CI = Confidence interval

CRP = C-reactive protein

CVD = Cardiovascular disease

DPP = Diabetes Prevention Program

ELISA = Enzyme-linked immunosorbent assay

ET-1 = Endothelin-1

FBG = Fasting blood glucose

FFA = Free fatty acids

GLP-1 = glucagon like peptide-1

HBP = High blood pressure

HDL = High density lipoprotein

Hs-CRP = High sensitive C-reactive protein

IDF = International Diabetes Federation

IGT = Impaired glucose tolerance

IL-6 = Interleukin-6

IR = Insulin resistance

LDL = Low-density lipoprotein

MAP = Mitogen activated protein

MetS = Metabolic syndrome

NCEP- ATP III = National Cholesterol Education Programme Adult Treatment Panel III

PAI-1 = Plasminogen activator inhibitor 1

PI3K = Phosphoinositide 3-kinase

RAP = Resonant acoustic profiling

RAS = Renin-angiotensin system

SAP = Serum amyloid P

SGLT-2 = Sodium glucose transporter-2

T2DM = Type 2 diabetes mellitus

TG = Triglycerides

TNFα = Tumor necrosis factor alpha

tPA = tissue plasminogen activator

TZD = Thiazolidinediones

VLDL = Very low-density lipoprotein

WC = Waist circumference

WHO = World Health Organization

CHAPTER ONE

INTRODUCTION, RATIONALE AND OBJECTIVES

1.1 Introduction:

Worldwide prevalence of metabolic syndrome (MetS) is widely diverse, ranges from <10% to 84%, depending on many factors including the region, urban or rural environment, composition (sex, age, race, and ethnicity) of the population studied, and the definition of the syndrome used (Kaur, 2014).

In Sudan, a recent study provides evidence for the high prevalence of MetS in Sudanese university students. It is varied depending on the definition used. The prevalence was highest by using International Diabetes Federation (IDF) criteria (8.4%) followed by National Cholesterol Education Programme Adult Treatment Panel III (NCEP ATP-III) (7.5%) and World Health Organization (WHO) (6.1%) (Sabir *et al*., 2016).

Recent meta-analysis of prospective cohort studies showed that MetS is associated with moderately increased cardiovascular disease (CVD) mortality and all-cause mortality in older populations.The pathogenesis of MetS involves both genetic and acquired factors which play a role in the final pathway of inflammation that leads to CVD. MetS has become increasingly relevant in recent times due to the exponential increase in obesity worldwide (Rochlani *et al*., 2017).

Another study reported that patients with MetS appear with 1.5- to three-fold higher risk for developing CVD and five times higher risk for Type 2 diabetes over the next 5–10 years compared with individuals without MetS (Kassi *et al*., 2011).

According to that, the prevalence of MetS is increasing globally, and thus predictive biomarkers are required for identification of MetS patients at an increased risk. Elevated levels of the inflammatory marker

high-sensitivity C-reactive protein (Hs-CRP) are associated with increased risk for CVD and diabetes mellitus, and the utility of Hs-CRP in predicting cardiovascular risk has been demonstrated in many studies. For example, in the Women's Health Study, Ridker et al found that baseline Hs-CRP cut points of 1.0, 1.0 to 3.0, and 3.0 mg/L improved prediction of relative risk of cardiovascular events (Haffner, 2006).

1.2 Rationale:

The prevalence of MetS is increasing globally. Accordingly the early screening is vital to predict and prevent the development of cardiovascular diseases and other consequences of MetS. Elevated levels of the inflammatory marker high-sensitivite C-reactive protein (Hs-CRP) are associated with increased risk for CVD and diabetes mellitus. In Sudan there are studies about the prevalence of MetS, but there is no information about Hs-CRP level in MetS patients. That is why we attempt to do this study to provide information that improves the monitoring, prognosis of risks associated with MetS and treatment program in patients.

1.3 Objectives:

1.3.1 General objective:

To assess serum High-sensitive C-reactive protein level among patients with metabolic syndrome.

1.3.2 Specific objectives:

1. To estimate High-sensitive C-reactive protein serum levels in study groups.
2. To compare between mean concentrations of High-sensitive C-reactive protein serum levels in case with control group.

3. To compare between mean concentrations of High-sensitive C-reactive protein serum levels according to variables subgroups (Age, Gender, BMI and Therapy usage).

4. To correlate between High-sensitive C-reactive protein serum levels and both of BMI and WC.

CHAPTER TWO

LITERATURE REVIEW

2.1 The metabolic syndrome:

Metabolic syndrome (MetS) is defined as a combination of impaired glucose metabolism, dyslipidemia, abdominal obesity, and elevated blood pressure. MetS had been defined slightly differently by various organizations. Three most popular definitions used for surveys and health care plan are:

WHO 1999: Presence of insulin resistance or glucose > 6.1 mmol/L (110 mg/dl), 2 h glucose > 7.8 mmol (140 mg/dl) (required) along with any two or more of the following: HDL cholesterol < 0.9 mmol/L (35 mg/dl) in men, < 1.0 mmol/L (40 mg/dl) in women, triglycerides > 1.7 mmol/L (150 mg/dl), waist/hip ratio > 0.9 (men) or > 0.85 (women) or BMI > 30 kg/m2 and/or blood pressure > 140/90 mmHg.

NCEP (National Cholesterol Education Program) ATP III **2005**: Presence of any three or more of the following: Blood glucose greater than 5.6 mmol/L (100 mg/dl) or drug treatment for elevated blood glucose, HDL cholesterol < 1.0 mmol/L (40 mg/dl) in men, < 1.3 mmol/L (50 mg/dl) in women or drug treatment for low HDL-C, blood triglycerides > 1.7 mmol/L (150 mg/dl) or drug treatment for elevated triglycerides, waist > 102 cm (men) or > 88 cm (women) and/or blood pressure > 130/85 mmHg or drug treatment for hypertension.

IDF (International Diabetes Federation) 2006: Waist > 94 cm (men) or > 80 cm (women) along with the presence of two or more of the following: Blood glucose greater than 5.6 mmol/L (100 mg/dl) or diagnosed diabetes, HDL cholesterol < 1.0 mmol/L (40 mg/dl) in men, < 1.3 mmol/L (50 mg/dl) in women or drug treatment for low HDL-C, blood triglycerides > 1.7 mmol/L (150 mg/dl) or drug treatment for

elevated triglycerides and/or blood pressure > 130/85 mmHg or drug treatment for hypertension (Saklayen, 2018).

According to the American Heart Association and the NHLBI the presence of three or more of any of the following parameters defines the presence of the metabolic syndrome in an individual. These parameters include:an elevated waist circumference. In women, 35 inches (88 cm); in men, 40 inches (102 cm), elevated triglyceride levels, i.e, 150 mg/dL, elevated fasting glucose, i.e, 100 mg/dL, reduced HDL-cholesterol. In women, 50 mg/dL; in men,40 mg/dL and/or elevated blood pressure, i.e, 130/85 mm Hg (Bishop *et al*., 2010).

Clinical measures	**WHO (1999)**	**ATP III (2005)**	**IDF (2006)**
Insulin resistance	IGT, IFG, T2DM, or lowered insulin Sensitivity **plus any 2 of the following**	**None, but any 3 of the following 5 features**	None
Body weight	Men: waist/hip ratio > 0.90 Women: waist/hip ratio > 0.85 and/or BMI> 30 kg/m2	WC > 102 cm in men or > 88 cm in women	Increased WC (population specific) **plus any 2 of the following**
Lipids	TGs ≥150 mg/dL and/or HDL-C <35 mg/dL in men or <39 mg/dL in women	TGs ≥150 mg/dL and HDL-C <40 mg/dL in men or <50 mg/dL in women	TGs ≥150 mg/dL or on TGs Rx. HDL-C <40 mg/dL in men or <50 mg/dL in women or on HDL-C Rx
Blood pressure	≥140/90 mmHg	≥ 130/85 mmHg	≥ 130/85 mmHg or on hypertension Rx
Glucose	IGT, IFG, or T2DM	> 110 mg/dL (includes diabetes)	≥100 mg/dL (includes diabetes)
Other	Microalbuminuria: Urinary excretion rate of > 20 mg/min or albumin: creatinine ratio of > 30 mg/g		

Table (2.1): Diagnostic criteria proposed for the clinical diagnosis of the MetS. BMI: body mass index; HDL-C: high density lipoprotein cholesterol; IFG: impaired fasting glucose; IGT: impaired glucose tolerance; Rx: receiving treatment; TGs: triglycerides; T2DM: type 2 diabetes mellitus; WC: waist circumference (Kaur, 2014).

2.1.1 Epidimiology of metabolic syndrome:

Worldwide prevalence of MetS ranges from <10% to as much as 84%, depending on the region, urban or rural environment, composition (sex, age, race, and ethnicity) of the population studied, and the definition of the syndromeused (Kaur, 2014).

Tanner and colleagues reported that: "The prevalence of MetS has beenestimated that 20% to 25% of adults worldwide have MetS. In 2000, it was estimated that 50 million US adults had MetS, and in studies of European countries,the prevalence of MetS has ranged widely but has exceeded50% in some studies and has routinely exceeded 20%. Similarly high rates have been noted in Asia and Latin America.For example, in a nationally representative sample of Chinese adults 35 to 74 years of age, Gu and colleagues reported a prevalence of MetS of 9.8% among men and 17.8% among women. Overall, these rates represent 71 million Chinese adults with MetS" (Tanner *et al*., 2012).

The prevalence estimates vary, based on the criteria used for the definition of MetS. For example, a national survey in Iran in 2007 showed prevalence of MetS was about 34.7% based on ATP III criteria, 37.4% based on IDF definition. In Tunisia, prevalence was 45.5% based on IDF criteria but 24.3% based on ATP III criteria. But in all the Middle Eastern countries, prevalence was much higher among women than men (Delavari*et al*., 2009).

Cameron et al. have concluded that the diet, differences in genetic background, smoking, levels of physical activity, family history of diabetes, and education all influence the prevalence of the MetS and its components (Cameron *et al*., 2004).

2.1.2 Pathophysiology of metabolic syndrome:

MetS is a state of chronic low grade inflammation as a consequence of complex interplay between genetic and environmental factors. These factors include: insulin resistance, visceral adiposity, atherogenic dyslipidemia, endothelial dysfunction, genetic susceptibility, elevated blood pressure, hypercoagulable state, and chronic stress.

2.1.2.1 Insulin resistance:

Insulin resistance (IR) is defined as a pathophysiological condition in which a normal insulin concentration does not adequately produce a normal insulin response in the peripheral target tissues such as adipose, muscle, and liver. Taking into consideration the main tissues targeted by insulin, insulin resistance in skeletal muscle results in a reduction in glycogen synthesis and glucose transport, while insulin resistance in the liver appears to lead to reduced effectiveness of insulin signalling pathways (McCracken et al., 2017).

In the early stages of insulin resistance, the pancreas compensates by increasing the secretion of insulin into the bloodstream in an attempt to overcome defects in peripheral insulin action. Although hyperinsulinemia may compensate for insulin resistance to some biological actions of insulin, it may cause an overexpression of insulin activity in some normally sensitive tissues, eventually decompensation will occur. This accentuation of some insulin actions coupled with a resistance to other actions of insulin results in the clinical manifestations of MetS.

Physiological insulin signalling occurs following the binding of insulin to the insulin receptor, a ligand-activated tyrosine kinase. Binding of insulin results in a tyrosine phosphorylation of downstream substrates and activation of two parallel pathways: the phosphoinositide 3-kinase (PI3K) pathway and the mitogen activated protein (MAP) kinase pathway. The PI3K-Akt pathway is affected, while, the MAP kinase pathway functions normally in insulin resistance. This leads to a change in the balance between these two parallel pathways. Inhibition of the PI3K-Akt pathway leads to a reduction in endothelial Nitric oxide production, resulting in an endothelial dysfunction, and a reduction in GLUT4 translocation, leading to a decreased skeletal muscle and fat glucose uptake. By contrast, the MAP kinase pathway is unaffected, so there is a continued endothelin-1 (ET-1) production, an expression of vascular cell adhesion molecules, and a mitogenic stimulus to vascular smooth muscle cells. In these ways, an insulin resistance leads to the vascular abnormalities that predispose to atherosclerosis. Although insulin resistant individuals need not be clinically obese, they nevertheless commonly have an abnormal fat distribution that is characterized by a predominant upper body fat. Regardless of the relative contributions of visceral fat and abdominal subcutaneous fat to insulin resistance, a pattern of abdominal (or upper body) obesity correlates more strongly with the insulin resistance and the MetS than does lower body obesity (Kaur, 2014).

2.1.2.2 Abdominal obesity:

Adipose tissue is a heterogeneous mix of adipocytes, preadipocytes, immune cells, and endothelium, which can respond rapidly and dynamically to alterations in nutrient excess through adipocytes hypertrophy and hyperplasia. Although not all overweight or obese individuals are metabolically disturbed, the majority are IR. Central obesity is thought to be an early step, as visceral adipose tissue secretes a

variety of bioactive substances termed adipocytokines, such as glycerol, free fatty acids (FFA), leptin, resistin, tumor necrosis factor alpha (TNFα), interleukin-6 (IL-6), and angiotensin II which induce IR, along with plasminogen activator inhibitor 1 (PAI-1), which is related to thrombogenic vascular diseases, and C-reactive protein (CRP) (Kassi *et al*., 2011).

Adipocytokines integrate the endocrine, autocrine, and paracrine signals to mediate the multiple processes including insulin sensitivity, oxidant stress, energy metabolism, blood coagulation, and inflmmatory responses which are thought to accelerate atherosclerosis, plaque rupture, and atherothrombosis. TNFα is a paracrine mediator in adipocytes and appears to act locally to reduce the insulin sensitivity of adipocytes. Evidence suggests that TNFα induces adipocytes apoptosis and promotes insulin resistance by the inhibition of the insulin receptor substrate 1 signalling pathway. The paracrine action would further tend to exacerbate the FFA release, inducing an atherogenic dyslipidemia. Plasma TNFα is positively associated with the body weight, WC, and triglycerides (TGs), while, a negative association exists between the plasma TNFα and High density lipoprotein–cholesterol (HDL-C) (Kaur, 2014).

Interleukin 6 (IL-6) is a cytokine produced by adipocytes and immune cells and has complex regulatory mechanisms. Production of IL-6 increases with increase in body fat and insulin resistance. It acts on the liver, bone marrow, and endothelium, leading to increased production of acute phase reactants in the liver, including C-reactive protein (CRP). Several studies have demonstrated a correlation between high CRP levels and the development of MetS, diabetes, and CVD. IL-6 also increases fibrinogen levels resulting in a prothrombotic state. It also promotes adhesion molecule expression by endothelial cells and activation of local renin-angiotensin system (RAS) pathways (Rochlani *et al*., 2017).

Elevated levels of CRP are associated with an increased WC, insulin resistance, BMI, and hyperglycemia, and are increased with the number of the MetS components. It is more likely to be elevated in obese insulin-resistant, but, not in obese insulin-sensitive subjects.

Other compounds produced by adipose tissue possibly implicated in the pathogenesis of MetS, are the non esterified free fatty acids (FFAs). In the presence of IR the process of FFAs mobilization from stored adipose tissue triglycerides is accelerated. In the liver, FFAs result (due to hepatic insulin resistance) in increased production of glucose and triglycerides and secretion of very low-density lipoprotein (VLDL), maintaining a vicious cycle. FFAs also reduce insulin sensitivity in muscle by inhibiting insulin-mediated glucose uptake and increase fibrinogen and PAI-1 production (Kassi *et al*., 2011).

PAI-1 is A serine protease inhibitor is secreted from intra-abdominal adipocytes, platelets, and the vascular endothelium. It exerts its effcts by inhibiting the tissue plasminogen activator (tPA) and thus is considered as a marker of an impaired firinolysis and atherothrombosis. Plasma PAI-1 levels are increased in abdominally obese subjects and inflmmatory states, thus, increasing the risk of an intravascular thrombus and adverse cardiovascular outcomes (Kaur, 2014).

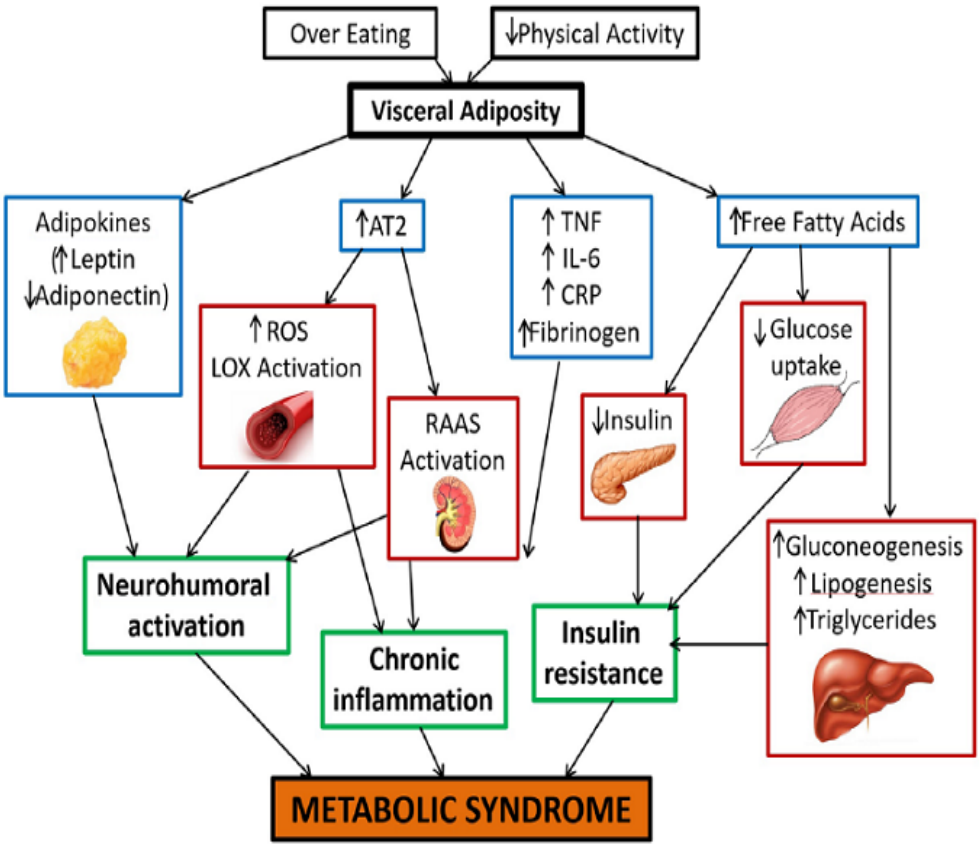


Figure (2.1): Pathophysiological mechanisms in metabolic syndrome. AT2, angiotensin II type 2 receptor; CRP, C-reactive protein; IL-6, interleukin 6; LOX, lectin-like oxidized low-density lipoprotein; RAAS, reninangiotensin-aldosterone system; ROS, reactive oxygen species; TNF, tumor necrosis factor (Rochlani *et al*., 2017).

2.1.2.3 Hypertension:

High blood pressure is an important constituent of the metabolic syndrome, but the underlying mechanisms for development of hypertension in the metabolic syndrome are very complicated and remain still obscure. Visceral/central obesity, insulin resistance, sympathetic overactivity, oxidative stress, endothelial dysfunction, activated renin-angiotensin system, increased inflammatory mediators, and obstructive sleep apnea have been suggested to be possible factors to develop hypertension in the metabolic syndrome (Yanai *et al*., 2008).

Studies suggest that both hyperglycemia and hyperinsulinemia activate the Renin angiotensin system (RAS) by increasing the expression of angiotensinogen, Angiotensin II (AT II), and the AT1 receptor, which, in concert, may contribute to the development of hypertension in patients with insulin resistance. There is also evidence that insulin resistance and hyperinsulinemia lead to SNS activation, and, as a result, the kidneys increase sodium reabsorption, the heart increases cardiac output, and arteries respond with vasoconstriction resulting in hypertension (Morse *et al*., 2005).

2.1.2.4 Dyslipidemia:

Most patients with metabolic syndrome have atherogenic dyslipidemia. The major component of atherogenic dyslipidemia is elevation of apo B-containing lipoproteins. These include low-density lipoprotein (LDL) and VLDL. Other components include elevated triglycerides and reduced high density lipoprotein cholesterol (HDL-C). Circulating lipoproteins filter into the arterial wall, where they are trapped, modified in various ways, and incorporated into macrophages to form lipid-laden foam cells. These cells dominate in the first stage of atherosclerosis, the fatty streak. As foam cells degrade they stimulate a connective tissue reaction, resulting in a fibrous plaque. This plaque usually covers a cholesterol rich core. In regions where the fibrous covering of this core is thin and enriched with macrophages, the plaque becomes unstable: rupture can occur, precipitating a thrombosis. Other risk factors of the metabolic syndrome accelerate this process. When plaque rupture occurs, the patient frequently suffers an acute cardiovascular event, e.g., myocardial infarction or stroke. Many clinical trials conclusively prove that intensive lowering of apo B containing lipoproteins reduces the risk for major cardiovascular events. Another component of atherogenic dyslipidemia is a low level of HDL-C. In epidemiologic studies, this abnormality

associates strongly with a higher risk for atherosclerotic cardiovascular disease (ASCVD) (Grundy, 2016).

2.1.2.5 Hypercoagulable State:

Individuals with obesity and metabolic syndrome appear to have multiple abnormalities in the haemostatic system that may well predispose to ASCVD. Among these are endothelial dysfunction, enhanced coagulation, impaired fibrinolysis, and platelet dysfunction. Specific abnormalities include elevated levels of inhibitor of plasminogen activator type 1(PAI-1), tissue factor, fibrinogen, and factor VIII activity (Grundy, 2016). These changes in the haemostatic system may favour the development of thrombosis. Hyperactivity of platelets and hypercoagulability favour platelet and fibrin deposits, and hypofibrinolysis due to the PAI-1 excess prevents their elimination. The increased PAI-1 expression that accompanies abdominal obesity is the most documented abnormality associated with the MetS. As PAI-1 could also be directly involved in the physiopathology of obesity, it could represent an original target for preventing both the thrombotic and metabolic risks (Alessi and Juhan-Vague, 2008).

2.1.3 Treatment of metabolic syndrome:

Clinical identification and management of patients with the MetS are important to begin efforts to adequately implement the treatments to reduce their risk of subsequent diseases. Effective preventive approaches include lifestyle changes, primarily weight loss, diet, and exercise, and the treatment involve the proper use of pharmacological agents to reduce the specific risk factors. Pharmacological treatment should be considered for those whose risk factors are not adequately reduced with the preventive measures and lifestyle changes. In 2001, NCEP ATP III recommended two major therapeutic goals in patients with MetS:

Treating underlying causes (overweight/obesity and physical inactivity) by intensifying weight management and increasing physical activity, and treating cardiovascular risk factors if they persist despite lifestyle modification.

2.1.3.1 Lifestyle Modifications:

(A) Weight reduction:

Four therapies can be used for weight reduction: calorie restriction (e.g., 500 kcal/d deficit), increased physical activity, behavioural modification, and, in appropriate patients, FDA-approved weight-reducing drugs. Several authors recommend a weight loss goal of 10% reduction in body weight in the first six months to a year and continued weight loss thereafter until BMI is less than 25. A weight loss of as small as 5–10% of body weight can significantly reduce TGs and increases HDL-C. Furthermore, both hypertensive individuals and individuals at risk of developing hypertension can see a significant reduction in the blood pressure with a modest weight loss. Fasting blood glucose, insulin, and haemoglobin A1c can also be decreased with a modest weight loss. During weight maintenance (i.e., energy balance), a regular exercise appears to play an important role in abdominal fat loss, and the prevention of weight regain in those who have successfully lost weight (Kaur, 2014).

(B) Diet:

Numerous trials reported that weight loss is beneficial for treating all components of MetS. The Finnish Diabetes Prevention Study showed that lifestyle intervention with modest weight loss significantly reduced the prevalence of MetS (odds ratio, 0.62; 95% CI, 0.40–0.95) compared with the control group. A 41% reduction in the incidence of MetS was seen

with the intensive lifestyle intervention arm of the Diabetes Prevention Program (DPP). For every kilogram of weight loss, the risk of diabetes development was decreased by 16%. Dietary carbohydrate can be divided into 2 categories: simple and complex. Complex carbohydrates should form the bulk of carbohydrate intake, whereas simple carbohydrates should be limited. A diet that is rich in complex, unrefined carbohydrates, high in fiber (14 g/1000 cal consumed daily), and low in added sugar (25% of caloric intake) is recommended for individuals with or at risk for MetS. These dietary changes contributed to weight loss, which was the primary predictor of a decrease in diabetes incidence in the study (Prasad *et al*., 2012).

A clear positive association has been shown between sodium intake and blood pressure, with excessive sodium intake associated with hypertension. Furthermore, a sodium restriction has also been associated with reduced CVD events, and congestive heart failure. Guidelines therefore recommend that a daily sodium intake should be restricted to no more than 65–100 mmol. In addition to sodium restriction, an increased potassium intake has also been shown to improve blood pressure, especially in the setting of high sodium intake. Guidelines have recommended the intake of foods enriched with potassium, such as fruits and vegetables, with a goal of 90–120 mmol of potassium per day (Appel *et al*., 2006).

The 2005 USDA dietary guidelines, primarily based on observational studies, recommend a reduction in saturated fat intake (less than 7% of caloric intake) and an increase in unsaturated fatty acids, specifically linoleic acid (5%–10% of caloric intake) and alpha-linolenic acid (0.7%–1.6% of caloric intake). These guidelines are also applicable for risk reduction of CVD. Both serum cholesterol and overall CVD risk have

been shown to be improved by this type of dietary fat intake. The Nurses' Health Study investigators reported that a 5% increase in saturated fat intake was associated with a 17% increase in coronary risk, whereas monounsaturated and polyunsaturated fat intakes were inversely related to CHD (Prasad *et al*., 2012).

(C) Physical Activity:

Physical inactivity is identified as the fourth leading risk factor for global mortality. Regular physical activity leads to enhanced energy consumption and is associated with reduced risk of prevalent diseases such as obesity, MetS, T2DM, CVD, cognitive impairment, depression, and osteoporosis. In MetS, the excess energy that is accumulated in adipose tissue and also stored ectopically in non adipose tissues like the liver will cause metabolic disturbances that lead to increases in BP, blood glucose, TGs, and inflammation. These metabolic alterations can be prevented or reduced if physical activity is performed daily. Any type of physical activity is better than inactivity, and increasing physical activity may also have substantial beneficial effects on personal well-being (Pérez-Martínez *et al*., 2017).

2.1.3.2 Pharmacological treatment:

In addition to lifestyle including weight loss, a targeted approach for control of individual components of the metabolic syndrome is often necessary. Because there are drugs proven effective in reducing specific components of metabolic syndrome, optimal pharmacological management must be individualized. First, because weight loss in patients with the metabolic syndrome increases insulin sensitivity, anti obesity drugs should be considered. In the USA the list includes, phentermine, extended release phentermine/topiratmate, lorcaserin, orlistat, and now

sustained release bupropion/naltrexone and the glucagon like peptide-1 (GLP-1) agonist liraglutide. Drugs such as thiazolidinediones (TZD) or metformin, which are insulin sensitizers, are recommended for insulin resistant patients. Metformin, rosiglitazone and pioglitazone are known to prevent type 2 diabetes in patients with and without the metabolic syndrome and pioglitazone also increases HDL-C and reduces triglycerides and hepatic steatosis. Lipid-lowering agents such as statins are drugs of choice for atherogenic dyslipidemia. Most but not all metabolic syndrome patients will be statin-eligible; moreover fibrates may reduce CVD events in patients who are hypertriglyceridemic and have reductions in HDL-C. New anti diabetic agents such as GLP-1 agonists and sodium glucose transporter-2 (SGLT-2) inhibitors result in some weight loss and are candidate drugs particularly in metabolic syndrome patients with or at risk for diabetes. RAS blockers are effective in decreasing blood pressure and other cardiovascular risk. Cilostazol, an anti platelet agent, is also known to improve atherogenic dyslipidemia and increase nitric oxide levels (Lim and Eckel, 2014).

2.2 High sensitive C-reactive protein:

C-reactive protein (CRP) is synthesized in the liver and is one of the first acute-phase proteins to rise in response to inflammatory disease. CRP received its name because it precipitates with the C substance, a polysaccharide of pneumococci. High-sensitivite CRP (Hs-CRP) is the same protein but is named for the newer, monoclonal antibody–based test methodologies that can detect CRP at levels below 1 mg/L (Bishop *et al*., 2010). Its link to CHD was reported more than 60 years later. CRP has been prodigiously investigated, largely facilitated by its relative stability as a frozen sample, long plasma half-life of 19 h, and ease of testing with a standardized assay (Yousuf *et al*., 2013).

In the past decade, high-sensitivity assays with rapid turnaround times for measurement have become available. High-sensitivity assay techniques such as immunonephelometry, immunoturbidimetry, high-sensitivite enzyme-linked immunosorbent assay (ELISA) and resonant acoustic profiling (RAP) can detect CRP with a sensitivity range of (0.01 to 10 mg/ l). These high-sensitivity assays help quantify low grades of systemic inflammation, in the absence of overt systemic inflammatory or immunologic disorders. The Hs-CRP assays have been standardized across several commercial platforms and can be accurately measured from fresh or frozen plasma. The Hs-CRP is the most widely evaluated biomarker in the quest for an ideal biomarker for global cardiovascular disease (CVD) risk prediction (Kamath *et al*., 2015).

2.2.1 Biochemistry of High sensitive C-reactive protein:

CRP belongs to the pentraxin family of calcium-dependent ligand-binding plasma proteins, the other member of which in humans is serum amyloid P component (SAP). The human CRP molecule is composed of five identical non glycosylated polypeptide subunits, each containing 206 amino acid residues. The protomers are non covalently associated in an annular configuration with cyclic pentameric symmetry (Figure 2.2). Each protomer has the characteristic "lectin fold," composed of a two-layered β sheet with flattened jellyroll topology. The ligand-binding site, composed of loops with two calcium ions bound 4 Å apart by protein side-chains, is located on the concave face. The other face carries a single α helix (Figure 2.2). The pentraxin family, named for its electron micrographic appearance from the Greek *penta* (five) *ragos* (berries), is highly conserved in evolution, with homologous proteins throughout the vertebrates and even in the phylogenetically distant arachnid, *Limulus polyphemus*, the horseshoe crab. SAP, named for its universal presence in

amyloid deposits, is a constitutive, non–acute-phase plasma glycoprotein in humans and all other species studied, except the mouse, in which it is the major acute-phase protein (Pepys and Hirschfield, 2003).

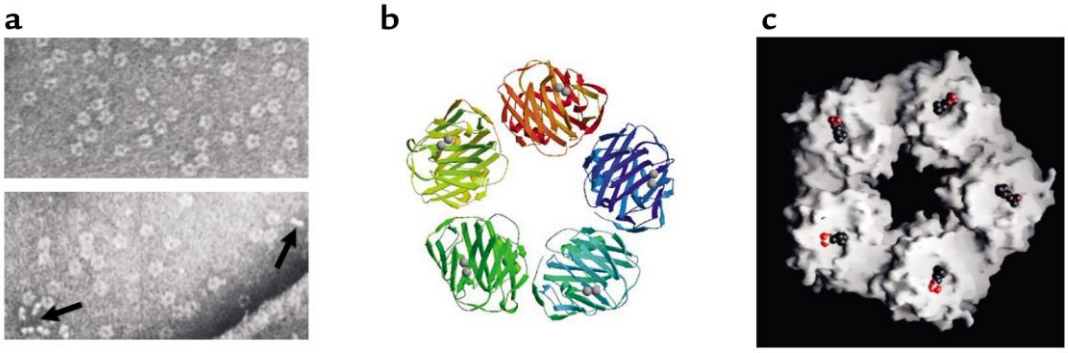


Figure (2.2): Molecular structure and morphology of human CRP. (**a**) Negatively stained electron micrograph showing the typical pentameric disc-likestructure face on and side-on (arrows). (**b**) Ribbon diagram of the crystal structure, showing the lectin fold and the two calcium atoms(spheres) in the ligand-binding site of each protomer. (**c**) Space filling model of the CRP molecule, showing a single phosphocholinemolecule located in the ligand-binding site of each protomer (Pepys and Hirschfield, 2003).

2.2.2 Clinical significance of High sensitive C-reactive protein:

2.2.2.1 Hs-CRP and Cardiovascular Diseases:

CRP is an acute-phase reactant and non specific marker of inflammation, produced predominantly in hepatocytes as a pentamer of identical subunits in response to several cytokines. Interleukin (IL)-6, one of the most potent drivers of CRP production, is released from activated leukocytes in response to infection or trauma and from vascular smooth muscle cells in response to atherosclerosis. CRP directly binds highly atherogenic oxidized low-density lipoprotein cholesterol (LDL-C) and is present within lipid-laden plaques. The possible mechanistic role of CRP in plaque deposition is highly complex, exerting proatherogenic effects in many cells involved in atherosclerosis. CRP may facilitate monocyte adhesion and transmigration into the vessel wall, a critical early step in the atherosclerotic process. Furthermore, M1 macrophage polarization,

catalyzed by CRP, is a proinflammatory trigger in plaque deposition, leading to macrophage infiltration of both adipose tissue and atherosclerotic lesions (Yousuf *et al*., 2013).

The utility of Hs-CRP in predicting cardiovascular risk has been demonstrated in many studies. For example, in the Women's Health Study, Ridker et al found that baseline Hs-CRP cut points of 1.0, 1.0 to 3.0, and 3.0 mg/L improved prediction of relative risk of cardiovascular events (according to the Framingham 10-year risk score) on multivariate analysis among 27,939 apparently healthy subjects. In an analysis among men in the primary prevention West of Scotland Coronary Prevention Study (WOSCOPS) statin trial, Hs-CRP level was associated with a significantly increased hazard ratio for CAD events of 1.36 (p 0.001) on univariate but not multivariate analysis. The presence of the metabolic syndrome using a modified NCEP ATP III definition (body mass index [BMI] criterion instead of waist circumference for abdominal obesity) was associated with a significantly increased hazard ratio for CAD of 1.76 (p 0.001) on univariate analysis and 1.30 (p 0.05) on multivariate analysis. For new-onset diabetes, the metabolic syndrome and Hs-CRP levels were associated with significantly increased risk ratios of 3.51 and 1.55 (both, p 0.001). CAD risk increased with the presence of increasing numbers of metabolic syndrome criteria (Haffner, 2006).

2.2.2.2 Role of Hs-CRP in disease intervention:

Several pharmacological agents that have demonstrated cardioprotective ability such as aspirin and statins will lower Hs-CRP and /or risk of CHD that is associated with increased Hs-CRP concentration. All statin drugs have been shown to reduce Hs-CRP, but interestingly the magnitude of LDL cholesterol reduction caused by statin therapy is minimally correlated with the magnitude of Hs-CRP reduction. Data from several large randomized trials suggest that the cardiovascular risk reduction

attributable to statin therapy may be most notable for those with increased Hs-CRP concentration at baseline. Thiazolidinediones (TZDs) also decrease inflammatory factors such as Hs-CRP and promote adipose tissue differentiation in subcutaneous adipose tissue regions, which increases the synthesis of adiponectin, thereby further reducing insulin resistance (Lim and Eckel, 2014).

CHAPTER THREE

MATERIALS AND METHODS

3.1 Materials:

3.1.1 Study design:

This is case control study.

3.1.2 Study area and period:

The study was carried out over 21 months (April 2019-December 2020) in Abdalla Khalil center and Mahdi center in Khartoum state, Sudan.

3.1.3 Ethical consideration:

This study was approved by the ethical committee of Federal Ministry of Health. Then an informed consent was obtained from participant (Appendix I), data was collected using questionnaire (Appendix II).

3.1.4 Study population:

This study included 33 metabolic syndrome patients (16 of them were males and 17 were females) and 33 healthy individuals as control group, age was matched in both groups, ranged from 28 to 75 years. Patients with metabolic syndrome were excluded from this study if they had chronic renal disease, thyroid disease, or pregnant women.

3.1.5 Sampling:

Three ml of venous blood was collected from each participant, placed in plain containers, Sample left clot at room temperature then serum was obtained after centrifuged for 3 minutes at 3000 RPM and analyzed immediately or kept until analysis, the participants underwent routine examinations that included the measurement of Height, weight, and BP. Weight and height were measured without shoes; BP was measured on the right upper arm and maintained at the level of the heart with participants in sitting position. BP was measured by trained and certified nurse's working in Hospital.

3.2 Methods:

3.2.1 Estimation of High sensitive C-reactive protein level:

Principle of the method:

Hs-CRP rapid test is based on Fluorescence immunoassay technology (FIA) uses a sandwich immune-detection method, when sample is added to the sample well of the test cartridge, the fluorescence-labelled detector CRP antibody binds to CRP antigen in blood specimen. As the sample mixture migrates on the nitrocellulose matrix of test strip by capillary action, the complexes of detector antibody and CRP are captured to CRP antibody that has been immobilized on test strip. Thus the more CRP antigens in blood specimen, the more complexes are accumulated on test strip. Signal intensity of fluorescence of detector antibody reflects amount of CRP captured and FIA Meter shows CRP concentrations in blood specimen. The default results unit of Hs-CRP is displayed as mg/L. (Appendix III).

Procedure of method:

The test was been operated at room temperature. ID Chip checked and inserted into the equipment, 5 μL of serum was drawn with a transfer pipette and add to the buffer tube, the specimen mixed well with buffer for 1 minute by inverting the tube. 75μL of sample mixture loaded onto the sample well of the test cartridge. The test cartridge inserted onto the test cartridge holder, 3 minutes later, the result was shown in the display. (Appendix III).

3.2.2 Quality control:

Each CRP rapid quantitative test cartridge contains internal control that satisfies routine quality control requirements. This internal control is performed each time a patient sample is tested. This control indicates that the test cartridge was inserted and read properly by FIA Meter. An

invalid result from the internal control causes an error message on FIA Meter indicating that the test should be repeated.

3.2.3 Calculation of BMI:

Body Mass Index was calculated by formula:

BMI =body mass divided by the square of body height (Bishop, 2010).

$$\frac{\text{Weight (kg)}}{\text{Height (m}^2\text{)}}$$

3.2.4 Statistical analysis:

Data were analyzed statistically by using the SPSS computer program (Version 20), the independent T test and chi-square were used for comparison, *P*-value (< 0.05) was considered significant.

CHAPTER FOUR

RESULTS

4. Results:

Table (4-1) shows clinical characteristics of study groups.

Table (4-2) shows that the level of Hs-CRP was significantly increased in metabolic syndrome patients compared to control group (20.70 ± 14.80 and 6.38 ± 1.38) respectively, with *P*-value (0.000).

Table (4-3) shows that there were no significant differences between MetS patients who were < 50 years and ≥50 years in Hs-CRP levels (22.75 ± 15.47 and 18.78 ± 14.34) respectively, with *P*-value (0.452).

Table (4-4) shows that there were no significant differences between males and females in Hs-CRP levels (17.46 ± 14.8 and 23.76 ± 14.56) respectively, with *P*-value (0.227).

Table (4-5) shows that there were no significant differences between overweight and obese MetS patients in Hs-CRP levels (20.4 ± 17.04 and 21.1 ± 12.1) respectively, with *P*-value (0.896).

Table (4-6) shows that the level of Hs-CRP was significantly decreased in patients who were using anti lipid drugs compared to patients who were not (13.5 ± 8.6 and 24.8 ± 16.2) respectively, with *P*-value (0.014).

Table (4-7) shows that there were no significant differences in Hs-CRP levels between patients who were using anti DM drugs and patients who were not (17.7 ± 12.9 and 24.2 ± 16.6) respectively, with *P*-value (0.216).

Table (4-8) shows that there were no significant differences in Hs-CRP levels between patients who were using anti HTN drugs and patients who were not (23.1 ± 17.1 and 18.1 ± 11.9) respectively, with *P*-value (0.351).

Table (4-9) shows that there was no correlation between the level of Hs-CRP and both of BMI and WC with (r = 0.079 and -0.118) respectively, but there was +ve, weak and significant correlation between BMI and WC with (r = 0.475).

Table (4-1): Clinical characteristics of study groups:

	Case (n = 33)	**Control** (n = 33)	***P*-value**
Gender	Males = 16 (48.5%) Females = 17 (51.5)	Males = 16 (48.5%) Females = 17 (51.5)	--
Age (years)	53.6 ± 11.5	52.2 ± 11.5	0.602
BMI (Kg/m^2)	29.1 ± 3.6	22.1 ± 4.5	0.000
WC (cm)	107.7 ± 10.3	84.1 ± 10.6	0.000
SBP (mm Hg)	128.2 ± 13.2	120.6 ± 2.4	0.001
DBP (mm Hg)	81.8 ± 5.6	80.6 ± 2.4	0.065

4.1 Comparison between metabolic syndrome patients and control for Hs-CRP:

Table (4-2): Comparison between metabolic syndrome patients and control for Hs-CRP:

Parameter	**Sample condition**		***P* - value**
	Case (n = 33)	**Control** (n = 33)	
Hs-CRP (mg/L)	20.70 ± 14.80	6.38 ± 1.38	0.000

4.2 Comparison between variables for Hs-CRP:

4.2.1 Comparison between Hs-CRP levels in MetS patients according to age groups:

Table (4-3): Levels of Hs-CRP for metabolic syndrome patients according to age groups.

Parameter	**Age**		***P* – value**
	< 50 years (n = 16)	**≥ 50 years** (n = 17)	
Hs-CRP (mg/L)	22.75 ± 15.47	18.78 ± 14.34	0.452

4.2.2 Comparison between Hs-CRP levels in MetS patients according to gender groups:

Table (4-3): Levels of Hs-CRP for metabolic syndrome patients according to gender groups.

Parameter	**Gender**		***P* – value**
	Male (n = 16)	**Female** (n = 17)	
Hs-CRP (mg/L)	17.46 ± 14.8	23.76 ± 14.56	0.227

4.2.3 Comparison between Hs-CRP levels in MetS patients according to BMI groups:

Table (4-5): Levels of Hs-CRP for metabolic syndrome patients according to BMI groups.

Parameter	**BMI**		***P* – value**
	Overweight (25-29.9 Kg/m^2) (n = 18)	Obese (≥ 30 Kg/m^2) (n = 15)	
Hs-CRP (mg/L)	20.4 ± 17.04	21.1 ± 12.1	0.896

4.2.4 Comparison between Hs-CRP levels in MetS patients according to therapy used:

Table (4-6): Levels of Hs-CRP for metabolic syndrome patients according to lipid-lowering drug usage.

Parameter	**Lipid-lowering drug**		***P* – value**
	Yes (n = 12)	**No** (n = 21)	
Hs-CRP (mg/L)	13.5 ± 8.6	24.8 ± 16.2	0.014

Table (4-7): Levels of Hs-CRP for metabolic syndrome patients according to glucose-lowering drug usage.

Parameter	**Glucose-lowering drug**		***P* – value**
	Yes (n = 18)	**No** (n = 15)	
Hs-CRP (mg/L)	17.7 ± 12.9	24.2 ± 16.6	0.216

Table (4-8): Levels of Hs-CRP for metabolic syndrome patients according to blood pressure-lowering drug usage.

Parameter	**Blood pressure-lowering drug**		***P* – value**
	Yes (n = 17)	**No** (n = 16)	
Hs-CRP (mg/L)	23.1 ± 17.1	18.1 ± 11.9	0.351

4.3 Correlation between Hs-CRP, BMI and WC of MetS patients:

Table (4-9): Correlation between Hs-CRP, BMI and WC of MetS patients:

Variables	r&P	Hs-CRP	BMI	WC
Hs-CRP	**r**	1	0.079	-0.118
	P	-	0.661	0.512
BMI	**r**	0.079	1	0.475
	P	0.661	-	0.005
WC	**r**	-0.118	0.475	1
	P	0.512	0.005	-

Figure (4.1): Correlation between Hs-CRP and BMI of MetS patients:

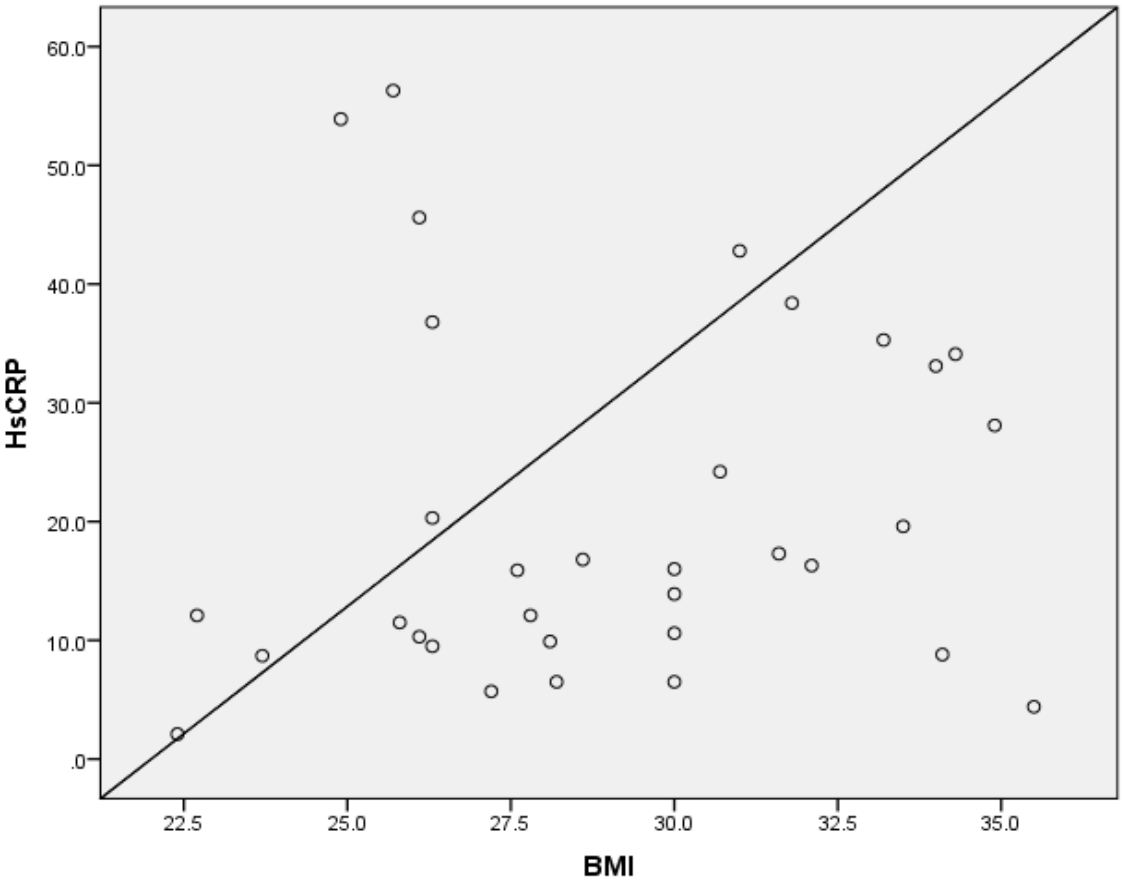


Figure (4.2): Correlation between Hs-CRP and WC of MetS patients:

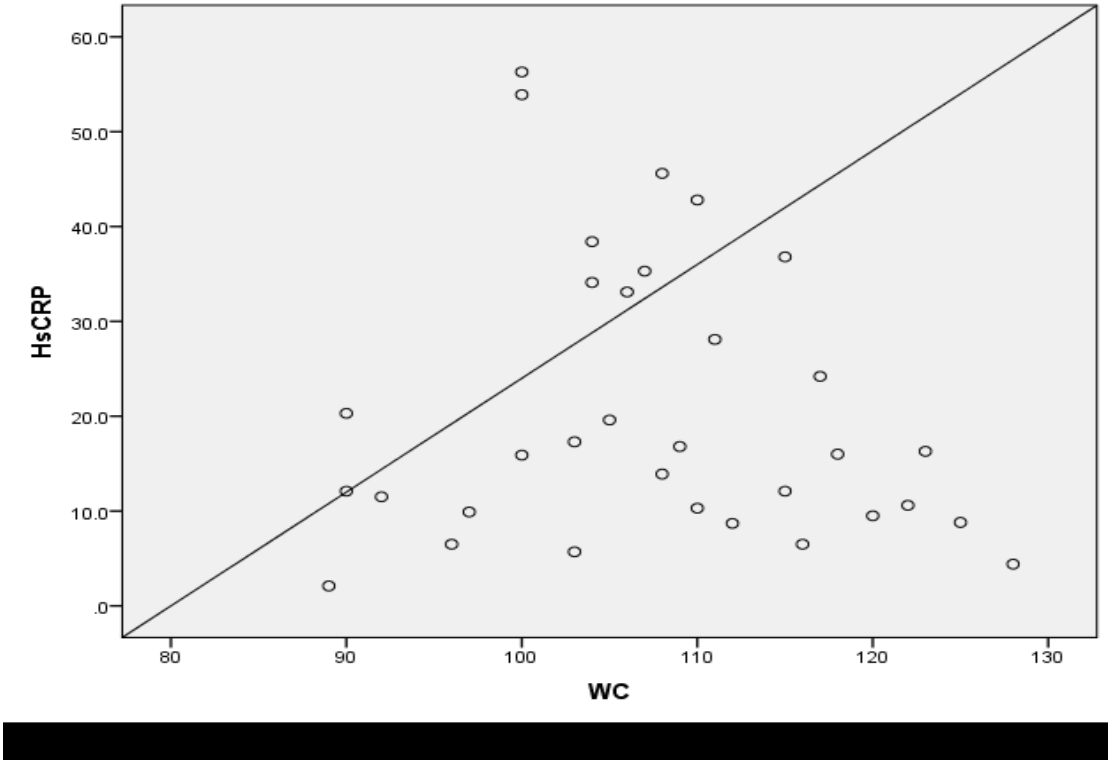
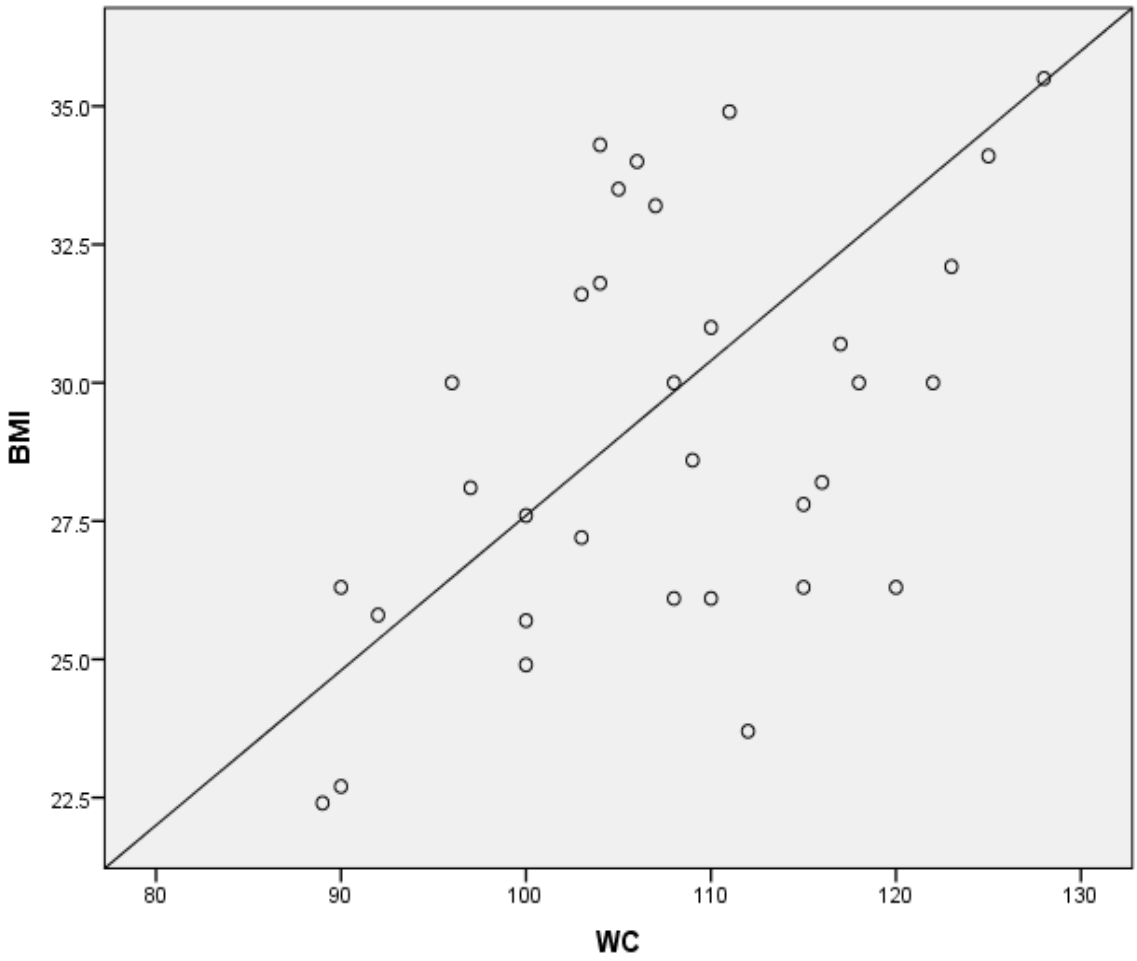


Figure (4.3): Correlation between BMI and WC of MetS patients:



CHAPTER FIVE

DISCUSSION, CONCLUSION AND RECOMMENDATIONS

5.1 Discussion:

MetS is a worldwide problem, its prevalence is widely diverse, ranges from <10% to 84%, and cardiovascular diseases are common complication of MetS (Kaur, 2014).

The result of present study showed that the level of Hs-CRP was significantly higher in MetS patients compared to control group with *P*-value (0.000), The cause of elevated levels of Hs-CRP seems to be due to persistent inflammation in MetS, this result agreed with recent study by (Mirhafez *et al*., 2016), who reported that the concentration of Hs-CRP was significantly increased in MetS patients compared to control group with *P*-value (0.001).

According to Age, Hs-CRP was insignificantly differ with increasing in age with *P*-value (0.452), this result disagreed with study by (Nadrowski et al., 2016), who reported that the mean of Hs-CRP increased significantly with increasing in age with *P*-value (0.007).

According to gender, Hs-CRP was insignificantly differ in males and females with *P*-value (0.227), this result agreed with recent study by (Alaei *et al*., 2018), who reported that the mean of Hs-CRP was insignificantly differ in females and males with *P*-value (0.140).

In concerning to BMI, Hs-CRP was insignificantly differ in overweight patients and obese patients with *P*-value (0.896), this result disagreed with study by (Guldiken *et al*., 2007), who reported that the mean of Hs-CRP increased significantly with increasing in BMI with *P*-value (0.010).

According to Therapy usage, Hs-CRP was significantly decreased in patients who were using lipid-lowerig drugs compared to patients who were not with *P*-value (0.014), this result was in agreement with recent study by (Kumar *et al*., 2019), who reported that the mean of Hs-CRP was significantly decreased in patients after using statins with *P*-value (0.000). This reduction in Hs-CRP level seems to be due to action of

lipid-lowering agents (such as statins and fibrates), which can diminish circulating levels of Hs-CRP (Lim and Eckel, 2014).

There were non-significant differences in Hs-CRP levels between patients who were using glucose-lowering drugs and patients who were not with *P*-value (0.216), this result was in accordance with previous study by (Abdulkadir and Thanoon, 2012), who reported that there were non-significant differences in Hs-CRP levels between patients before and after metformin therapy with *P*-value (> 0.05).

There were non-significant differences in Hs-CRP levels between patients who were using blood pressure-lowering drug drugs and patients who were not with *P*-value (0.351), this result disagreed with recent study by (Ali *et al*., 2018), who reported that Hs-CRP levels was significantly decreased in patients who were using regular anti HTN drugs compared to patients who were not with *P*-value (0.018). The difference was non-significant probably due to the limited sample size of this study.

In concerning to correlation, showed that there was no correlation between the level of Hs-CRP and both of BMI and WC with (r = 0.079 and -0.118) respectively.

The correlation between BMI and WC was +ve, weak and significant correlation with (r = 0.475), this result agreed with study by (Gierach *et al*., 2014), who reported that WC was found to be significantly correlated with BMI with (r= 0.780).

5.2 Conclusion:

MetS patients had increased Hs-CRP levels which might be associated with increased risk of CVD.

5.3 Recommendations:

- The serum concentrations of the Hs-CRP could be done routinely for MetS patients.
- Additional biochemical parameters could be considered, to increase the specificity of Hs-CRP as predictive marker, such as: fibrinogen, angiopoietin-like protein 8 and leptin/Adiponectin ratio.

References

References:

Abdulkadir A. and Thanoon I. (2012). "Comparative effects of Glibenclamide and metformin on C-reactive protein and Oxidant/Antioxidant status in patients with type II diabetes mellitus", Sultan Qaboos Univ Med J; **12**(1): 55-61.

Alaei Faradonbeh N., Nikaeen F., Akbari M., Almasi N., Vakhshoori M. (2018). "Cardiovascular disease risk prediction among Iranian patients with diabetes mellitus in Isfahan Province, Iran, in 2014, by using Framingham risk score, atherosclerotic cardiovascular disease risk score, and high-sensitive C-reactive protein", ARYA Atheroscler; **14**(4): 163-168.

Alessi M.-C., and Juhan-Vague I. (2008). "Metabolic syndrome, haemostasis and thrombosis", Thrombosis and Haemostasis; **99**(06): 995–1000.

Ali E., Abd Elrhman M. and Mahmoud M. (2018). "High-sensitivity C-reactive protein as a potential marker for hypertension", Al-Azhar Assiut Med J; **18**: 13-20.

Appel L. J., Brands M. W., Daniels S. R., Karanja N., Elmer, P. J., and Sacks F. M. (2006). "Dietary Approaches to Prevent and Treat Hypertension: A Scientific Statement From the American Heart Association", Hypertension; **47**(2): 296–308.

Bishop M. L., Fody E. P. and Schoeff L. E. (2010). "Clinical Chemistry: Techniques, Principles, Correlations", 6th Edition, **1**: 652.

Cameron A. J., Shaw J. E. and Zimmet P. Z. (2004). "The metabolicsyndrome: prevalence in worldwide populations",

Endocrinology and Metabolism Clinics of North America; **33**(2): 351.

Dayal D., Jain H.,attri S., BHarti B. and BHalla A. (2014). "Relationship of High Sensitivity C-Reactive Protein Levels to Anthropometric and other Metabolic Parameters in Indian Children with Simple Overweight and Obesity", Journal of Clinical and Diagnostic Research; **8**(8): 5-8.

Delavari A., Forouzanfar M. H., Alikhani S., Sharifian A., and Kelishadi R. (2009). "First nationwide study of the prevalence of the metabolic syndrome and optimal cutoff points of waist circumference in the Middle East: the national survey of risk factors for noncommunicable diseases of Iran", Diabetes Care; **32**(6):1092–1097.

Gierach M., Gierach J., Ewertowska M., Arndt A., and Junik R. (2014). "Correlation between Body Mass Index and Waist Circumference in Patients with Metabolic Syndrome", ISRN Endocrinology; **1**: 1-6.

Grundy S. M. (2016). "Metabolic syndrome update", Trends in Cardiovascular Medicine; **26**(4): 364–373.

Guldiken S., Demir M., Arikan E., Turgut B., Azcan S., Gerenli M. and Tugrul A. (2007). "The levels of circulating markers of atherosclerosis and inflammation in subjects with different degrees of body mass index: Soluble CD40 ligand and high-sensitivity C-reactive protein", Thrombosis Research; **119**(1): 79—84.

Haffner S. M. (2006). “The Metabolic Syndrome: Inflammation, Diabetes Mellitus, and Cardiovascular Disease”, The American Journal of Cardiology; **97**(2): 3–11.

Kamath D. Y., Xavier D., Sigamani A. and Pais P. (2015). "High sensitivity C-reactive protein (hsCRP) & cardiovascular disease: An Indian perspective", Indian J Med Res; **142**(3): 261–268.

Kassi E., Pervanidou P., Kaltsas G. and Chrousos G. (2011). "Metabolic syndrome: definitions and controversies", BMC Med; **9**: 48.

Kaur J. (2014). “A Comprehensive Review on Metabolic Syndrome”, Cardiology Research and Practice; **1**: 1–21.

Kumar B., Shah M., Kumar R., Kumar J. and Memon A. (2019) "Comparison of Atorvastatin and Rosuvastatin in Reduction of Inflammatory Biomarkers in Patients with Acute Coronary Syndrome", Cureus: **11**(6): 4898.

Lim S., and Eckel R. H. (2014). "Pharmacological treatment and therapeutic perspectives of metabolic syndrome", Reviews in Endocrine and Metabolic Disorders; **15**(4): 329–341.

McCracken E., Monaghan M., and Sreenivasan S. (2017). "Pathophysiology of the metabolic syndrome", Clinics in Dermatology; **9**: 4.

Mirhafez S., Ebrahimi M., Saberi Karimian M., Avan A., Tayefi M., Heidari-Bakavoli A., Parizadeh M., Moohebati M., Azarpazhooh M., Esmaily H., Nematy M., Safarian M., Ferns G. and Ghayour-Mobarhan M. (2016). "Serum high-sensitivity C-reactive protein as a biomarker in patients with metabolic

syndrome: evidence-based study with 7284 subjects", European Journal of Clinical Nutrition; **70**(1): 1298-1304.

Morse S. A., Zhang R., Thakur V., & Reisin E. (2005). "Hypertension and the Metabolic Syndrome", The American Journal of the Medical Sciences; **330**(6): 303–310.

Nadrowski P., Chudek J., Skrzypek M., Puzianowska M, Mossakowska M., Wiecek A., Zdrojewski T., Grodzicki T., Kozakiewicz K., (2016). "Associations between cardiovascular disease risk factors and IL-6 and hsCRP levels in the elderly", Experimental Gerontology; **10**: 1-28.

Pepys M. B., and Hirschfield G. M. (2003). "C-reactive protein: a critical update", Journal of Clinical Investigation; **111**(12): 1805–1812.

Pérez-Martínez P., Mikhailidis D. P., Athyros V. G., Bullo M., Couture P., Covas M. I., and López-Miranda J. (2017). "Lifestyle recommendations for the prevention and management of metabolic syndrome: an international panel recommendation", Nutrition Reviews; **75**(5): 307–326.

Prasad H., Ryan D. A., Celzo M. F., and Stapleton D. (2012). "Metabolic Syndrome: Definition and Therapeutic Implications", Postgraduate Medicine; **124**(1): 21–30.

Rochlani Y., Pothineni N.V., Kovelamudi S. and Mehta J.L.(2017). "Metabolic syndrome: pathophysiology, management, and modulation by natural compounds", Therapeutic Advances in Cardiovascular Disease; **11**(8): 215–225.

Sabir F.M., Hassan D.A., and Elamin M.I. (2016). "Prevalence of Metabolic Syndrome among YoungSudanese University Students Using ThreeDifferent Criteria of WHO, IDF and NCEP-ATPIII", Pediatr Neonatal Nurs Open Access; **2**(2): doi: http://dx.doi.org/10.16966/2470-0983.112.

Saklayen M. G. (2018). "The Global Epidemic of the Metabolic Syndrome", Current hypertension reports; **20**(2): 12.

Yanai H., Tomono Y., Ito K., Furutani N., Yoshida H., & Tada N. (2008). "The underlying mechanisms for development of hypertension in the metabolic syndrome", Nutrition Journal; **7**(1): 10.

Yousuf O., Blaha J.M. And Budoff J. M. (2013). "High-Sensitivity C-Reactive Protein and Cardiovascular Disease", Journal of the American College of Cardiology; **62**(5): 397–408.

Appendicies

Appendix I

Informed Consent

الموافقة المستنيرة

أنا الباحث محمد الطيب عمر ابراهيم من كلية علوم المختبرات الطبية قسم الكيمياء السريرية، جامعة السودان للعلوم والتكنولوجيا، أقوم بدراسة علمية بحثية لنيل درجة الماجستير بعنوان تأثير متلازمة التمثيل الغذائي على مستوى البروتين سي التفاعلي عالي الحساسية لدى مرضى متلازمة التمثيل الغذائي في ولاية الخرطوم.

Assessment of Serum Level of High Sensitive C-Reactive Protein among Patients with Metabolic Syndrome - Khartoum State

متلازمة التمثيل الغذائي هي مجموعة من الحالات المرضية – زيادة ضغط الدم وارتفاع نسبة السكر في الدم والدهون الزائدة في الجسم حول منطقة الخصر وتغيير مستويات الكلسترول والدهون الثلاثية بشكل غير طبيعي- التي تحدث معا لتزيد من خطر الإصابة بأمراض القلب والسكتة الدماغية والسكري. إن الإصابة بحالة واحدة فقط من هذه الحالات المرضية لا تشير إلى الإصابة بمتلازمة التمثيل الغذائي.

لقد قمنا باختيارك لتشارك في هذا البحث العلمي لأنك تنطبق عليك كل شروط الشخص الذي يمكن أن يكون ضمن المشمولين للدراسة (شخص بالغ مصاب بداء السكري، وضغط الدم، ليس مصاب بمرض الكلى أو الغدة الدرقية).

خلال الدراسة سنقوم بسحب 3 ملم من الدم لإجراء تحليل البروتين سي التفاعلي عالي الحساسية، وأيضا سأقوم بأخذ معلومات عنك وعن المرض، علما بأن سحب العينة قد يؤدي إلى حدوث بعض الألم، وأيضا قد يؤدي إلى ظهور تورم في منطقة الحقن، قد يتفشى خلال ساعات وظهور كدمات زرقاء وسوف نعمل على تفادي كل هذه المضاعفات.

بعد الموافقة منك وأخذ المعلومات الكافية والإجابة عن الأسئلة المطلوبة في البحث، سوف يتم أخذ العينة والعمل على تحليلها، وسيتم إخبارك بالنتائج التي حصلنا عليها خلال فترة من الزمن، والتي سوف تكون في سرية تامة ولن يطلع عليها أحد غير العاملين في البحث لدون التعرف على محددات الهوية الشخصية، علما بأن إشتراكك سيكون طواعية ويمكنك الإنسحاب من الإشتراك في أي وقت تشاء دون خسائر، ونحيكم علما بأنه لن تكون هناك أي عوائد مالية نتيجة لإشتراكك في البحث، ويمكنك الإتصال على الباحث في الرقم ..... في أي وقت للمزيد من المعلومات خلال فترة البحث.

إقرار المشارك في البحث:

لقد اطلعت على المعلومات الحالية والتي تم شرحها لي وأتيح لي طرح الأسئلة عنها كيفما شئت، وقد تلقيت الإجابات الوافية عن كل الأسئلة، وأنا أقر بالموافقة على المشاركة طواعية في هذه الدراسة وأعلم بحقي في التوقف عن المشاركة في أي وقت دون أن يؤثر ذلك على حقوقي الأخرى والإستفادة من نتائج الدراسة.

توقيع المشارك:.....

ت – المشارك:.....

توقيع الباحث:.....

التأريخ:.....

بسم الله الرحمن الرحيم

Sudan University for Sciences and Technology

College of Graduate Studies

Assessment of Serum Level of High Sensitive C-Reactive Protein among Patients with Metabolic Syndrome - Khartoum State 2019

No: ……

Age: ( ) years.

Gender: Male ☐ Female ☐

Weight/Kg: ……

Height/cm: ……

BMI: ……...

Waist circumference/cm: ……

Systolic blood pressure/mmHg: ……

Diastolic blood pressure/mmHg: ……

Type of treatment:

Metformin ☐ Other antidiabetic ☐

Antihypertensive ☐ Statin ☐ Fibrates ☐

Family history of metabolic syndrome:

Yes ☐ No ☐

Laboratory Investigations

- Serum Hs-CRP result: …… mg/L

-Date …… -Tele No ……

-Signature ……..