SERUM LIPID PROFILE AND URIC ACID COMPARSION AMONG RHEUMATOID ARTHRITIS PATIENTS AND APPARENTLY HEALTH CONTROLS AT WORABE COMPHERENSIVE SPECIALIZED HOSPITAL, WORABE, SOUTHERN ETHIOPIA

BY: RESHAD NUREDIN (MSc. CANDIDATE)

A THESIS SUBMITTED TO SCHOOL OF MEDICAL LABORATORY SCIENCES, FACULTY OF HEALTH SCIENCES, INSTITUTE OF HEALTH, JIMMA UNIVERSITY, IN PARTIAL FULFILLMENT OF THE REQUIREMENT OF MASTERS OF SCIENCE DEGREE IN CLINICAL LABORATORY SCIENCE SPECIALITY IN CLINICAL CHEMISTRY

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JIMMA, ETHIOPIA

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By: - RESHAD NUREDIN (MSc. CANDIDATE)

ADVISORS: -

1. Mr. SHIFERAW BEKELE (MSc, Asis. Prof of CLS),
2. Mr. MISGANA BEKELE (MSc in CLS),
3. Mr. FANTA OBSA (MSc in CLS)

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JIMMA, ETHIOPIA

ABSTRACT

Background*: Rheumatoid arthritis is a chronic autoimmune condition that mostly damages joints. Warm, swollen, and painful joints are frequently the result, especially in the hands and wrists. Due to a higher risk of cardiovascular disease, people with rheumatoid arthritis have higher rates of morbidity and death than the general population. An invivo experiment demonstrated that uric acid has a potent ability as a proinflammatory molecule derived from dying cells and These evidences suggest the possibility that uric acid could contribute to systemic inflammatory conditions such as rheumatoid arthritis.*

Objective: *To compare serum lipid profile and uric acid among rheumatoid arthritis patients and apparently health individual controls at Worabe comprehensive specialized hospital from December 1, 2021, to April 1, 2022.*

Material and Methods*: An institution-based Comparative cross-sectional study design was carried out to compare serum lipid profile and uric acid among patients with rheumatoid arthritis and apparently healthy individuals at worabe comprehensive specialized hospital. A total of 228 Study subjects were included and selected by using convenient sampling techniques. Questionnaires were used to collect socio-demographic and clinical data. Anthropometric data were collected by an experienced nurse. A 5mL blood specimen was collected on a serum separator tube. The lipid profile and uric acid were analyzed by the Roche Cobas c 311 automated clinical chemistry analyzer. The Statistical Package for Social Science (SPSS) version 25 software was used to analyze the data.*

Results: *The results of our study showed that there was significant elevation of the median and interquartile range of TC, TG, LDL-C, TC/HDL-C, LDL-C/HDL and SUA but a lower value of HDL-C was seen among rheumatoid arthritis patients than controls (P value < 0.05). SUA had a significant negative correlation with HDL-C (ρ= -0.449), but a significant positive correlation with TC (ρ= 0.692), TG (ρ= 0.555), LDL-C (ρ= 0.695), and TC/HDL-C (ρ= 0.621).*

Conclusion: *The result of this study showed that significant elevation of TC, TG, LDL-C, TC/HDL, LDL/HDL and SUA but, lowered value of HDL-C was seen among RA patients than controls. So, it is possible to conclude that dyslipidemia was seen among RA patients than controls.*

Key words*: Rheumatoid arthritis, serum lipid profiles, cardiovascular risk, serum uric acid*

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TABLE OF CONTENTS

LIST OF TABLES AND FIGURES

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

ACR	American College of Rheumatology
BMI	Body Mass Index
CVD	Cardiovascular Diseases
DMARD	Disease-Modifying Anti-Rheumatic Drugs
EULAR	European League Against Rheumatism
HDL-C	High Density Lipoprotein Cholesterol
LDL-C	Low Density Lipoprotein Cholesterol
RA	Rheumatoid Arthritis
SOPs	Standard Operating Procedures
SUA	Serum Uric Acid
TC	Total Cholesterol
TG	Triglyceride
WCSH	Worabe Comprehensive Specialized Hospital

OPERATIONAL DEFINITION

Dyslipidemia: An abnormal lipid profile is defined in accordance with the US National Cholesterol Education Program, adult treatment panel III (NCEP-ATP III) guidelines as TC ≥ 200mg/dl, HDL-C < 40mg/dl, LDL-C ≥ 130mg/dl, TG ≥ 150 mg/dl.

Lipid Profile: A panel of blood tests for measurement of lipids in serum, such as TC, TG, HDL-C, and LDL-C.

Rheumatoid arthritis: is a chronic autoimmune disease that causes both systemic and articular inflammation above the age of 18.

CHAPTER ONE

1. INTRODUCTION

1.1. BACKGROUND

Rheumatoid arthritis (RA) is a chronic autoimmune disease that causes both systemic and articular inflammation, especially of the small joints of the hands and feet(1). Warm, swollen, and painful joints are typical symptoms, and the condition may also impact the skin, eyes, lungs, heart, nerves, blood, and other body organs. (2). Often, symptoms come on gradually over weeks to months(3). Rheumatoid arthritis's specific cause is unknown; however, it is thought to be a result of a mix of hereditary and environmental factors. The underlying mechanism involves the body's immune system attacking the joints. This results in inflammation and thickening of the joint capsule, with damage to the underlying bone and cartilage(2).

When autoimmune tissue destruction occurs in RA, it manifests as synovitis, an inflammation of the joint capsule made up of the synovial membrane, synovial fluid, and the corresponding bones (4). Multiple dendritic cell (DC) subtypes, T cells, macrophages, B cells, neutrophils, fibroblasts, and osteoclasts work in concert to start and maintain this joint inflammation. The constant immune cell activation causes a chronic inflammatory state in the joint and swelling of the synovial membrane because the RA-specific autoantigens that are present everywhere cannot be completely cleared. Affected patients experience pain and joint swelling as a result of this self-perpetuating inflammation(5). The periarticular bone at the cartilage-bone junction is invaded by the pannus, an enlargement of the synovial membrane caused by the ongoing chronic inflammation in the arthritic joint, which in turn causes bone erosion and cartilage degeneration(4).

Higher rates of morbidity and death are observed in rheumatoid arthritis (RA) patients than in the general population, which is largely attributable to an elevated risk of cardiovascular disease (CVD) in RA patients(6, 7). The elevated risk of cardiovascular disease (CVD) is thought to be associated with coronary atherosclerosis(8, 9) and may be caused either directly by chronic inflammation or indirectly by inactivity and RA medication use(10).

The activity of the RA disease appears to alter lipid levels. High-density lipoproteins (HDL) may undergo changes due to inflammation, which could reduce their capacity to remove cholesterol from atherosclerotic plaque and weaken their antioxidant properties(11).

RA is linked to unfavorable lipid profiles, such as a higher total cholesterol/HDL-cholesterol ratio or the so-called atherogenic lipoprotein phenotype, which is characterized by low HDL, high triglyceride levels, and elevated levels of tiny dense LDL. In addition to LDL-cholesterol levels, such pro-atherogenic lipid profiles are associated with higher cardiovascular risk(12, 13).

Atherosclerotic plaque development occurs in steps that are triggered by lipid deposition and oxidation in the subendothelial region, activation of leukocytes and endothelial cells, and ultimately thrombosis(14).

All lipoproteins carrying apolipoprotein (apo) B can pass through altered endothelial cells with damaged tight junctions and enter the subendothelial region(15, 16). These lipoproteins can be ingested by macrophages, which will then transform them into foam cells. Chylomicrons and their remnants do not require alteration, whereas LDL needs to be oxidized (oxLDL) before it can cause foam cell development(17). There is evidence that tumor necrosis factor alpha (TNF) can oxidize LDL directly(18), and it has been observed that oxLDL levels are higher in patients with RA(19). In addition, elevated levels of oxLDL have been connected to elevated RA disease activity. Lp-PLA2 (lipoprotein associated phospholipase A2) catalyzes the oxidation of LDL, although it is unclear how much this process contributes to the development of atherosclerosis in RA given that Lp-PLA2 levels are decreased in RA(20), whereas higher Lp-PLA2 activity has been linked to CVD(21).

Lp(a), a pro-atherogenic lipoprotein made up of an apo(a) and an LDL-like particle, can become oxidized and elicit a similar immunological response as oxLDL. Due to its similarity with plasminogen, apo(a) encourages thrombosis and prevents fibrinolysis(22). Inflammation has also been linked to a rise in Lp(a), however the evidence is conflicting. Lp(a) may be disproportionately high in RA and is an independent risk factor for CVD(23).

Inflammation plays a crucial role in the onset of both atherosclerosis and RA(24). The synovial tissue produces pro-inflammatory cytokines like TNF and interleukin-6 (IL-6), which are crucial to the pathophysiology of RA and the growth of atherosclerosis(25) An increase in monocyte

activation and cytokine release is one of TNF's effects(25, 26). Atherosclerotic plaque development, T and B cell proliferation, neutrophil recruitment, and tissue damage in RA are all impacted by IL-6, which also increases the likelihood of plaque rupture. In RA, the levels of IL-6 and TNF are both increased(25).

All these processes are brought about by the production of pro-inflammatory cytokines from synovial tissue, which cause systemic inflammation. In addition to inducing inflammatory changes in adipose tissue, these circulating cytokines may also boost the production of adipokines, which would further exacerbate systemic inflammation(15, 27).

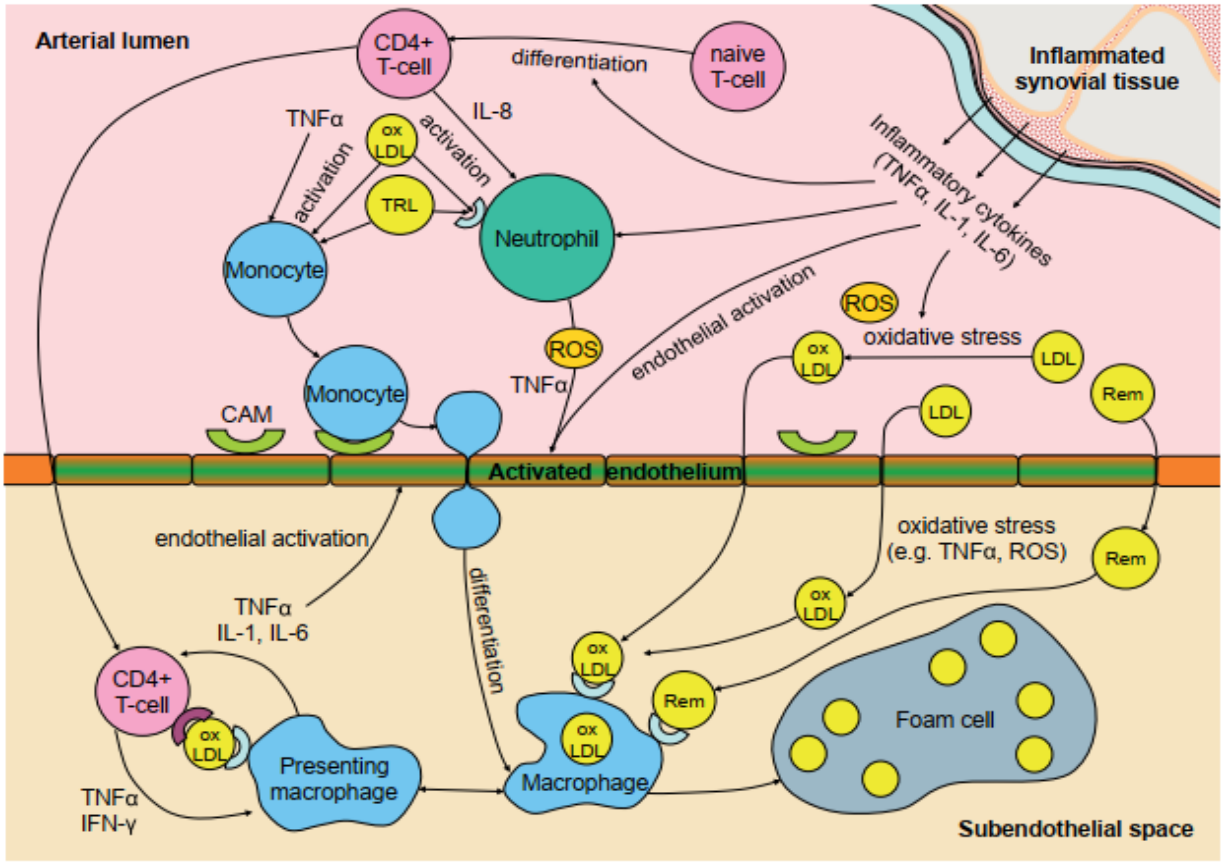

Figure 1. The inflammation-driven atherogenicity of rheumatoid arthritis (RA) (14)

The symptoms of the patient, examination findings, risk factor evaluation, joint evaluation by ultrasound sonography, and evaluation of laboratory indicators, such as high levels of CRP and ESR in the blood and the detection of RA-specific autoantibodies, are all used to make the diagnosis of RA(28).

Prior to the 1990s, RA usually caused disability, unemployment, and higher mortality rates, but more recent therapeutic choices have made the condition more tolerable. Here, there has been significant advancement in the creation of disease-modifying anti-rheumatic medicines (DMARDs), which work to reduce inflammation and stop future deterioration of the joints. The DMARDs that are currently on the market are divided into three categories: (a) conventional synthetic DMARDs (methotrexate, hydrochloroquine, and sulfadiazine); (b) targeted synthetic DMARDs (pan-JAK-and JAK1/2-inhibitors); and (c) biologic DMARDs (tumor necrosis factor (TNF)-inhibitors, TNF-receptor (R) inhibitors, IL-6 inhibitors, and IL-6R inhibitors, B cell depleting antibodies, and inhibitors of co-stimulatory molecules)(28).

Uric acid is produced in humans through the metabolism both of endogenous and exogenous purines(29). Growing evidence indicates that serum uric acid may have a major impact on inflammatory responses. It has been demonstrated that soluble uric acid induces the production of monocyte chemoattractant protein-1 (MCP-1) by vascular smooth muscle cells via nuclear factor-B (NF-B) and p38 mitogen-activated protein kinase (MAPK)(30). Uric acid stimulates the production of proinflammatory cytokines in human mononuclear cells, including interleukin-1 (IL-1), IL-6, and tumor necrosis factor (TNF-α)(31). An invivo experiment demonstrated that uric acid has a potent ability as a proinflammatory molecule derived from dying cells(32). In a population-based study, the relationship between serum uric acid levels and IL-6, C-reactive protein (CRP), and TNF-alpha was discovered to be strong(33, 34). These evidences suggest the possibility that uric acid could contribute to systemic inflammatory conditions such as rheumatoid arthritis (RA).

The potential correlation of SUA with lipid profile in rheumatoid arthritis patients has not been any detail in Ethiopia as well as Africa. So, the purpose of conducting this study is to assess the concentration of serum lipid profiles and serum uric acid in rheumatoid arthritis patients.

1.2. STATEMENT OF THE PROBLEM

Rheumatoid arthritis is an autoimmune condition that affects people all over the world and is a major source of mortality and morbidity. It is one of the illnesses with the biggest social impact. The prevalence of RA was 0.24% and remained unchanged between 1990 and 2010, according to the Global Burden of Disease 2010 study and clinical and epidemiological research. According to this study, between 1990 and 2010 there was an increase, going from 3.3 million to 4.8 million. This was caused by population growth and an aging population(35).

It is one of the leading causes of chronic morbidity in industrialized nations(36). It is an autoimmune condition that mostly impacts the little joints in the hands, wrists, and feet. If left untreated, it can result in severe cartilage erosion, which can deform the body and leave one disabled. Common symptoms include stiffness and discomfort, but prolonged illness is also linked to psychological issues like depression(37).

Rheumatoid arthritis (RA) is the prototypical chronic inflammatory rheumatic disease and affects 5 to 10 people per 1000 in industrialized nations. Delaying treatment causes severe RA, which results in physical impairment, a poor quality of life, and early death(38). It should be noted that from 1990 to 2017, RA caused 3.4 million disability-adjusted life years worldwide(39). Additionally, RA patients have up to 50% more documented deaths than the overall population(40-42). To mitigate the RA impact on the population, early detection and treatment of prevalent cases are paramount.

Rheumatoid arthritis (RA) is a chronic, systemic, inflammatory condition that mostly affects joints and has an unknown cause. The overall minimum prevalence of RA in the United Kingdom is 1.16 % in women and 0.44 percent in males(43). Recent, mounting evidence associates RA with excessive cardiovascular morbidity and mortality, including acute coronary syndromes (ACS), heart failure and coronary death(44).

It is likely that the situation in Africa is similar given that RA has a large impact on the burden of disease in many other emerging regions of South America and Asia(36). Despite the lack of data on RA incidence in Africa, efforts have been made to increase awareness and support for rheumatic disorders. In acknowledgment of the significant burden that this category of illness

represents, the African League of Associations for Rheumatology (AFLAR) was established in 1989 (45).

AFLAR has been tasked with documenting and improving the care of rheumatoid patients in Africa in conjunction with the Community Oriented Program for Central Control of Rheumatic Diseases (COPCORD), which was established by WHO to record the burden caused by rheumatic disorders through population surveys(37). Contrary to this initiative, only Egypt and Tunisia have COPCORD centers throughout Africa, and only one study by AFLAR on the incidence of RA has been published(37). Rheumatic illness now receives relatively little attention in Africa, with only one rheumatologist caring for Kenya's 16 million people and thirty for South Africa's 40 million as recently as 2003(46).

There is a research gap on rheumatoid arthritis and cardiovascular diseases in Ethiopia and throughout Africa. However, study results from Tikur Anbessa Specialized Hospital evaluated lipid profiles and high sensitivity C-reactive protein(47). Additionally, there is a dearth of information regarding the general lipid profile and uric acid levels of rheumatoid arthritis patients. As a result, this study evaluated the uric acid and lipid profiles of rheumatoid arthritis patients in the study area to close the knowledge gap and provide recommendations to reduce CVD-related morbidities and fatalities.

1.3. SIGNIFICANCE OF THE STUDY

Knowing the comparison of serum lipid profile and uric acid among patients with rheumatoid arthritis at Worabe comprehensive specialized hospital is important for the government to set appropriate strategies to control and manage dyslipidemia and inflammation and it will serve as a baseline for future research. It helps policymakers pay attention to serum lipid profiles and serum uric acid of rheumatoid arthritis patients to control dyslipidemia and inflammation.

CHAPTER TWO

2. LITERATURE REVIEW

Rheumatoid arthritis is caused by genetic and environmental factors, which result in immune dysregulation and inflammation. It is an inflammatory disorder characterized by chronic inflammation of synovial joints associated with proliferation of synovial cells and infiltration of activated immune inflammatory cells, which leads to progressive destruction of cartilage and bone. Complications of RA are mainly associated with the risk of cardiovascular disease. The morbidity and mortality of cardiovascular diseases increase in RA patients due to factors including, hypercholesterolemia and elevated systemic inflammation(47).

Cross-sectional investigations were carried out between 2013 and 2014 at the Jinnah Postgraduate Medical Center's (JPMC) "Rheumatology Clinic" in Karachi, Pakistan. The study included 200 Rheumatoid Arthritis (RA) patients in total. All patients had their fasting lipid profiles (LDL, HDL, and total cholesterol) completed. A total of 107 individuals (53.5%) had dyslipidemia, and the most prevalent abnormality a low HDL was seen in 83 (41.5%) of these patients(48).

A case-control study with 140 participants was carried out in India between 2016 and 2017 with the aim of comparing dyslipidemia and hyperuricemia. According to the study, the case group's uric acid levels were substantially higher than those of the controls (6.40± 1.27 vs. 4.89± 0.21 mg/dl). In patients with dyslipidemia, the levels of total cholesterol (TC), triglycerides (TAGs), LDL-C, and VLDL-C significantly increased. However, when compared to controls, levels of HDL-C in patients significantly decreased. Significantly positive relationships existed between uric acid and TC, TAGs, LDL-C, and VLDL-C. However, among patients, uric acid significantly correlated negatively with HDL-C. The findings of this study suggest that patients with dyslipidemia have a stronger correlation with hyperuricemia than do healthy subjects(49).

In Tehran, a cross-sectional study on RA patients was carried out between September 2014 and September 2015. Triglycerides (TG), total cholesterol (Chol), low-density lipoprotein (LDL), and high-density lipoprotein (HDL) were all examined in the laboratory. A diagnosis of dyslipidemia was made in 65.3 percent of the 150 individuals that were evaluated, with a mean

age of 54.9 ± 16.8 years. Dyslipidemia was more common in menopausal females and in patients with more advanced illness(50).

180 study volunteers were involved in the study that was carried out in Iraq between September and November 2020. Between the ages of 20 and 60, the study's control group consists of 60 healthy people and 120 female rheumatoid arthritis sufferers. The findings showed that lipid profile parameters in rheumatoid arthritis were significantly higher than the control values(51).

The Rheumatology Clinic of Tikur Anbessa Specialized Hospital carried out a hospital-based cross-sectional study. The study compared risk factors (dyslipidemia and inflammatory state) between people with RA as a case group and people who appeared to be in good health as a control group. The findings of this investigation showed that RA patients' mean±SD TC, TC/HDL, LDL/HDL, and HDL-C values were significantly higher than those of controls. When compared to controls, RA patients had significantly higher mean and SD levels of the inflammatory marker high-sensitivity C-reactive protein (hsCRP). A hsCRP and HDL-C had a significant inverse relationship, but TC/HDL-C and LDL/HDL-C had a significant positive relationship(47).

CHAPTER THREE

3. OBJECTIVES OF THE STUDY

3.1. GENERAL OBJECTIVE

- To assess and compare of serum lipid profile and uric acid among rheumatoid arthritis patients and apparently health controls at Worabe comprehensive specialized hospital from December 1, 2021, to April 1, 2022.

3.2. SPECIFIC OBJECTIVE

- ✓ To assess the level of serum lipid profiles among RA patients and controls from December 1, 2021, to April 1, 2022.
- ✓ To measure serum uric acid among RA patients and controls groups from December 1, 2021, to April 1, 2022.
- ✓ To determine the correlation of lipid profile parameters with serum uric acid levels of rheumatoid arthritis patients from December 1, 2021, to April 1, 2022.

CHAPTER FOUR

4. METHOD AND MATERIALS

4.1. STUDY AREA AND SETTING

The study was carried out in the Silte Zone, Southern Ethiopia, at Worabe Comprehensive Specialized Hospital. According to the 2007 census conducted by the Central Statistical Agency, the total population of the Silte Zone was 1,250,398 of whom 612,696 were men and 637,702 were women. Of these, 78,525 or 6.28% were urban inhabitants. Worabe town is the capital city of the Silte Zone, which is located 174 kilometers south of Addis Ababa. In Worabe town, there is one hospital and one health center. WCSH was established in 2014 in G.C. The hospital has more than 200 beds. Medical wards have 37 beds, pediatric wards have 38 beds, surgical wards have 38 beds, gynecology wards have 18 beds, eye clinics have 10 beds, NICU has 10 beds, and AICU has 4 beds.

4.2. STUDY DESIGN AND PERIOD

An institution-based comparative cross-sectional study was conducted to determine the level of serum lipid profile and uric acid among rheumatoid arthritis patients at Worabe comprehensive specialized hospital from December 1, 2021, to April 1, 2022.

4.3. POPULATION

4.3.1. SOURCE POPULATION

The source populations of this study were all rheumatoid arthritis patients and for the control group, all apparently healthy individuals at Worabe comprehensive specialized hospital.

4.3.2. STUDY POPULATION

All rheumatoid arthritis patients who consent and fulfill our inclusion criteria, and for the control group, all apparently healthy individuals who consent during the study period.

4.4. INCLUSION AND EXCLUSION CRITERIA

4.4.1. INCLUSION CRITERIA

Study subjects included in this study were those who fulfilled the 2010 revised criteria of the American Rheumatism Association for the classification of rheumatoid arthritis (joint involvement, abnormalities in CRP and ESR, presence of RA-specific autoantibodies, and overall symptom duration) (52) at the age of 18 years old and above and who volunteered to participate in this study. For the control group, apparently healthy individuals who take care of patients and staff members, aged 18 years or older who volunteered to participate in this study were included.

4.4.2. EXCLUSION CRITERIA

Patients with rheumatoid arthritis with the following conditions were excluded from the study: RA patients with clinically evident gout, diabetes mellitus, hypertension, thyroid disorders, liver disorders, and cancer.

4.5. SAMPLE SIZE DETERMINATION AND SAMPLING TECHNIQUES

The Sample size was calculated using the double population proportion formula for comparison of two populations' mean. The total sample size of this study was calculated by OpenEpi, Version 3 open-source calculator software and the mean and the standard deviation of TC were taken from a study done at Tikur Anbessa specialized hospital(47). Two-sided confidence Intervals (95%), 80% power and desired ratio of sample size for cases to controls gave a total sample size of 152, but most of the studies done before were not equal proportion, so to increase the sample size we could double the control group and the total sample was 228(76 cases and 152 controls).

Comparison of two means (sample size in each group)

$n = (S1^2 + S2^2)(Z\alpha/2 + Z\beta)^2 / (m1 - m2)^2$

Where,

M1and s12-are mean and variance of group 1 respectively.

m2and s22- mean and variance of group 2 respectively and,

Power = 80% and the level of significance = 5%

$N= (1631.35^2 + 937.584^2)\ 10.5/\ (179.64 – 163.35)^2 = 152$

76 participants were involved in the case group and 152 in the control group, for a total of 228 individuals. A convenient sampling technique was used to recruit individuals who fulfill the inclusion criteria until the required sample size is reached.

4.5.1. SAMPLING TECHNIQUE

A convenient sampling technique was used to collect all the available data during the study period consecutively.

4.6. STUDY VARIABLES

4.6.1. DEPENDENT VARIABLE

Serum lipid profiles (TC, TG, HDL-C and LDL-C) and Serum uric acid

4.6.2. INDEPENDENT VARIABLE

Age

Sex

Educational status

Marital status

Residence

BMI

Smoking

Physical exercise

Alcohol usage

Types of oil used

Types of occupation

4.7. DATA COLLECTION INSTRUMENTS

- Cobas c 311 clinical chemistry analyzer
- Controls (both normal and pathological)
- Lipid profile and uric acid reagents
- Serum separator tube
- Cuvette
- Centrifuge
- Disposable syringe
- 70% alcohol
- Cotton
- Glove
- Tourniquet
- Safety box
- Sample rack
- Weight scale
- stadiometer
- Questionnaires
- Pen

4.8. DATA COLLECTION TECHNIQUES

RA patients and apparently healthy individuals were interviewed to obtain socio-demographic data by using an interviewer-administered questionnaires and anthropometric parameters like; height and weight were measured by clinical nurses. Five ml of blood was withdrawn from the study participants, for laboratory analysis

4.8.1. MEASUREMENT OF WEIGHTAND HEIGHT

Anthropometric measurements were measured by professional nurses using a standardized protocol.

For the measurement of weight, the participant asked to remove his/her shoes and any bulky clothing and the participant's arms should be hanging freely by the sides of the body, with palms facing the thighs. The participant should hold his/her head up, and face forward.

The height was measured by instructing each subject's feet pointed outward: legs straight and knees together; arms at sides; head, shoulder blades, buttocks, and heels touching the measurement surface; looking straight ahead; and shoulders relaxed.

Body Mass Index (BMI) was calculated as weight in kilograms divided by height in meter squared (kg/m^2), and the results were recorded.

4.8.3. BLOOD SPECIMEN COLLECTION AND ANALYSIS

Five milliliters of random venous blood were collected from the antecubital veins of each study subject. Secondly, the blood specimen was allowed to stay for 30 min for clot formation. Then, the specimen would be centrifuged at 3000 revolutions per minute (rpm) and the serum would be separated from the whole blood. The Cobas c 311 analyzer was then used to measure the serum lipid profile and serum uric acid.

TOTAL CHOLESTEROL: Cholesterol was measured enzymatically in serum in a series of coupled reactions that hydrolyzed cholesterol esters and oxidize the 3-OH group of cholesterol. One of reaction products, H2O2 is measured quantitatively in per oxidase catalyzed reaction that produces color. Absorbance was measured at 500nm. The color intensity is proportional to cholesterol concentration. Desirable or normal cholesterol levels was considered to be those below 200mg/dl.

TRIGLYCERIDE: The method was based on the enzymatic hydrolysis of serum triglyceride to glycerol and fatty acids (FFA) by lipoprotein lipase (LPL). The glycerol is phosphorylated by Adenosine triphosphate (ATP) in the presence of glycerol kinase (GK) to form glycerol-3-phosphate (G-3-P) and adenosine diphosphate (ADP). G-3P is then oxidized by glycerophosphate oxidase (GSO) to form dihydroxyacetone phosphate (DHAP) and hydrogen peroxide. A red colored product is formed by peroxidase (POD) catalyzed coupling of 4-amnioantipyrine (4- AA) and phenol with hydrogen peroxide(H2O2), the optical density was read at 540 nm of which is proportional to the concentration of triglyceride in the sample.

HIGH DENSITY-LIPOPROTEIN CHOLESTEROL (HDL-C): Homogeneous enzymatic colorimetric test. In the presence of magnesium ions, dextran sulfate selectively forms water-soluble complexes with LDL, VLDL, and chylomicrons which are resistant to PEG-modified enzymes. The cholesterol concentration of HDL-cholesterol is determined enzymatically by cholesterol esterase and cholesterol oxidase coupled with PEG to the amino groups (approx. 40%). Cholesterol esters are broken down quantitatively into free cholesterol and fatty acids by cholesterol esterase. In the presence of oxygen, cholesterol is oxidized by cholesterol oxidase to delta 4-cholestenone and hydrogen peroxide.

In the presence of peroxidase, the hydrogen peroxide generated reacts with 4-amino-antipyrine and HSDA to form a purple-blue dye. The color intensity of the blue dye formed is directly proportional to the HDL-cholesterol concentration. It is determined by measuring the increase in absorbance photometrically.

LOW DENSITY LIPOPROTEIN CHOLESTEROL (LDL-C): This automated method for the direct determination of LDL-cholesterol takes advantage of the selective micellary solubilization of LDL-cholesterol by a nonionic detergent and the interaction of a sugar compound and lipoproteins (VLDL and chylomicrons). When a detergent is included in the enzymatic method for cholesterol determination (cholesterol esterase, cholesterol oxidase coupling reaction), the relative reactivities of cholesterol in the lipoprotein fractions increase in this order: HDL < chylomicrons < VLDL < LDL. In the presence of Mg++, a sugar compound markedly reduces the enzymatic reaction of the cholesterol measurement in VLDL and chylomicrons. The combination of a sugar compound with detergent enables the selective determination of LDL-cholesterol in serum.

SERUM URIC ACID: determination was based on enzymatic colorimetric test. Uricase cleaves uric acid to form allantoin and hydrogen peroxide. In the presence of peroxidase, 4-aminophenazone is oxidized by hydrogen peroxide to a quinone-diimine dye. The red color intensity of the quinone-diimine formed is directly proportional to the uric acid concentration.

Reagent Preparation (*Reagents are ready to use; no preparation required)

•**R1** reagent. Phosphate buffer: 0.05 mol/L, pH 7.8; TOOS: 7 mmol/L; fatty alcohol polyglycol ether: 4.8 %; ascorbate oxidase (EC 1.10.3.3; zucchini) ≥ 83.5 µkat/L (25 °C); stabilizers

•**R2** reagent. Phosphate buffer: 0.1 mol/L, pH 7.8; potassium hexacyanoferrate (II): 0.3 mmol/L; 4-aminophenazone ≥ 3 mmol/L; uricase (EC 1.7.3.3; Arthrobacter protophormiae) ≥ 83.4 µkat/L (25 °C); peroxidase (POD) (EC 1.11.1.7; horseradish) ≥50 µkat/L (25 °C); stabilizers

4.9. DATA QUALITY MANAGEMENT

The qualities of data were maintained through training given for data collectors on the objective of the study and how to retain confidentiality and privacy of the study subjects. The English version of the questionnaire was translated first to Amharic and then to Siltigna (local language). The collected data were checked for completeness and accuracy and corrected on a daily basis. Data were labelled before entry and edited appropriately by the principal investigator and entered into statistical computer package using statistical package for social science (SPSS version 25 and checked for missing data by principal investigator. Standardized operating procedures (SOPs) were followed during specimen collection and processing. All reagents were checked for their expiration dates and prepared according to the manufacturer's instructions. During the blood analysis, internal quality control materials were performed before study participants' samples were analyzed. Study participant samples were run after internal quality control; both normal and pathological controls were passed.

4.10. STATISTICAL ANALYSIS

Data were entered and analyzed with SPSS version 25. The data were tested for normality with the help of Shapiro-Wilk tests(if p- value is less than 0.05, the data are not normally distributed). All continuous variables with non-normally distributed values were expressed as a median (interquartile range) and frequencies or percentages for categorical variables. The significance median differences between cases and controls were determined by the Mann-Whitney U test. A Spearman's coefficient correlation was performed to determine the correlation between lipid profiles and serum uric acid. p-value ≤ 0.05 was considered statistically significant.

4.11. ETHICAL CONSIDERATION

Before starting data collection, ethical clearance letters were obtained from the institutional review board at Jimma University, Institute of Health, and permission to conduct the research was obtained from the clinical director of the WCSH. The English version of the questionnaire was translated first to Amharic and then to Siltigna (the local language). The selected respondents were well informed about the purpose of the study, and information was collected only after verbal or written consent from each participant was obtained.

Confidentiality was assured throughout the study period. To ensure confidentiality of data, study participants were identified using codes, and unauthorized persons had no access to the collected data.

4.12. PLAN FOR UTILIZATION AND DISSEMINATION OF RESULTS

The findings of this study will be submitted to Jimma University, Institute of Health, Faculty of Health Sciences, School of Medical Laboratory Sciences, and Worabe Comprehensive Specialized Hospital. The study, on completion, could serve as a reference material and a baseline for researchers and experts for further study. The result will also be disseminated through publication in peer-reviewed local and international journals and through presenting it in relevant seminars and workshops.

CHAPTER FIVE

5. RESULTS

5.1. SOCIO-DEMOGRAPHIC CHARACTERISTICS

A total of 228 subjects were involved in this study making hundred percent response rate (100%). Among the total study participants, 76 were RA patients and 152 were controls. This study enrolled 61(80.3%) females and 15(19.7%) males with rheumatoid arthritis patients. The majority of the rheumatoid arthritis patients were found within the age group of 38–57 years. Regarding marital status, most of the patients in the study were married 39 (51.3%), followed by single 15(19.7%). Nearly half of arthritis patients, 37 (48.7%), were unable to read and write. Most of the patients in the study were rural residents 51(67.1%), and most of them were 40(52.6%) housewives, followed by farmers 17(22.4%).

Table1. Socio- demographic characteristics of the study participants from December 1, 2021, to April 1, 2022 at WCSH, Worabe, Southern Ethiopia (n=228).

Characteristics		Cases (n = 76) N (%)	Control (n=152) N (%)	Total(n=228) N (%)
Age	18-37	21(27.6%)	44(28.9%)	65(28.5%)
	38-57	48(63.2%)	94(61.8%)	142(62.3%)
	58-77	7(9.2%)	14(9.2%)	21(9.2%)
Sex	Male	15(19.7%)	53(34.9%)	68(29.8%)
	Female	61(80.3%)	99(65.1%)	160(70.2%)
Residence area	Urban	25(32.9%)	59(38.8%)	84(36.8%)
	Rural	51(67.1%)	93(61.2%)	144(63.2%)
Marital status	Single	15(19.7%)	28(18.4%)	43(18.9%)

	Married	39(51.3%)	97(63.8%)	136(59.6%)
	Divorced	12(15.8%)	19(12.5%)	31(13.6%)
	Widowed	10(13.2%)	8(5.3%)	18(7.9%)
Educational status	Illiterate	37(48.7%)	22(14.5%)	59(25.9%)
	Write and read	14(18.4%)	20(13.2%)	34(14.9%)
	Complete primary school	11(14.5%)	36(23.7%)	47(20.6%)
	Complete secondary school	10(13.1%)	32(21%)	42(18.4%)
	Complete college or University	4(5.3%)	42(27.6%)	46(20.2%)
Regular Physical exercise	Yes	10(13.1%)	37(24.3%)	47(20.6%)
	No	66(86.9%)	115(75.7%)	181(79.4%)
Smoking habit	Yes	3(3.9%)	0	3(1.3%)
	No	73(96.1%)	152(100%)	225(98.7%)
Khat Chewing	Yes	26(34.2%)	19(12.5%)	45(19.7%)
	No	50(65.8%)	133(87.5%)	183(80.3%)
Alcohol drinking	No	74(97.4%)	151(99.3%)	225(98.7%)
	Non habitual	1(1.3%)	1(0.7%)	2(0.9%)
	Habitual	1(1.3%)	0	1(0.4%)
Type of oil used	Palm cooking	20(26.3%)	11(7.2%)	31(13.6%)
	Olive	54(71.1%)	141(92.8%)	195(85.5%)

	Other	2(2.6%)	0	2(0.9%)
Types of occupation	Farmer	17(22.4%)	21(13.8%)	38(16.7%)
	House wife	40(52.6%)	43(28.3%)	83(36.4%)
	Merchant	15(19.7%)	47(30.9%)	62(27.2%)
	Civil servant	2(2.6%)	41(27%)	43(18.9%)
	Industry worker	2(2.6%)	0	2(0.9)

(N = number of categorical demographic data, % = percent value of frequency of data)

5.2. CATEGORICAL VALUES OF SERUM LIPID PROFILES AND URIC ACIDS IN RHEUMATOID ARTHRITIS PATIENTS AND CONTROL GROUPS

Out of the total 76 RA patients, 19(25.0%), 26(34.2%), 12(15.8%), and 9(11.8%) were above the baseline value of the NCETPATP-III guideline for TC, TG, LDL-C, and UA respectively, but the value of HDL-C 15(19.7%) was below the baseline value of the NCETPATP-III guideline. These patients had dyslipidemia. On the other hand, out of 152 controls, 6(3.9%) and 9(5.9%) were above the baseline value of the NCETPATP-III guideline for TC and TG respectively, but 8(5.3%) HDL-C value was below the baseline value of the NCETPATP-III guideline

We compared the level of dyslipidemia between case and control groups according to the baseline value. The patients had 13(21.1%), 17(28.3%), and 12(15.8%) TC, TG, and LDL-C higher values than controls, respectively. But RA patients showed 7(14.4 %) lowered HDL-C values compared to the control group.

Table 2. Categorical values of some demographic and serum lipid profiles and uric acid in rheumatoid arthritis patients and control groups from December 1, 2021 to April 1, 2022 at WCSH.

Variables		Cases (n= 76) N (%)	Control (n= 152) N (%)	Total(n=228) N (%)
BMI (Kg/m2)	< 18.5	2(2.6%)	5(3.3%)	7(3.1%)
	18.5-24.9	66(86.8%)	135(88.8%)	201(88.2%)
	25-29.9	8(10.5%)	12(7.9%)	20(8.8%)
TC (mg/dl)	< 200	57(75.0%)	146(96.1%)	203(89.0%)
	≥ 200	19(25.0%)	6(3.9%)	25(11.0%)
TG (mg/dl)	< 150	50(65.8%)	143(94.1%)	193(84.6%)
	≥ 150	26(34.2%)	9(5.9%)	35(15.4%)
HDL-C (mg/dl)	≥ 40	61(80.3%)	144(94.7%)	205(89.9%)
	< 40	15(19.7%)	8(5.3%)	23(10.1%)
LDL-C (mg/dl)	< 130	64(84.2%)	152(100.0%)	216(94.7%)
	≥ 130	12(15.8%)	0	12(5.3%)
UA (mg/dl)	< 7.2	67(88.2%)	152(100%)	221(96.1%)
	≥ 7.2	9(11.8%)	0	9(3.9%)
TC/HDL-C	< 5	69(90.8%)	150(98.7%)	219(96.1%)
	≥5	7(9.8%)	2(1.7%)	9(3.9%)

5.3. COMPARISON OF SERUM LIPID PROFILES AND URIC ACID AMONG CASE AND CONTROL GROUP

RA patients had significantly higher median and interquartile range values, 174(162-200), 145(138-157), 100(93-120) and 5.8(5-6.7) of TC, TG, LDL-C and UA than control group values, 155(147.5-166.5), 125(115-138), 77(70-86) and 3.8(3.5-4.6) of TC, TG, LDL-C and UA respectively with p-value (<0.001). But the median and interquartile range value of HDL-C in case, 43(40.5-48) was significantly lower than control group value50(47.54) respectively with p-value (<0.001). From patients' medical chart, out of 76 total RA patients, 57 were positive for Rheumatoid factor test. The sensitivity was 75%.

Table 3. Comparison of serum lipid profiles and uric acid among case and control group from December 1, 2021 to April 1, 2022 at WCSH, Worabe, Southern Ethiopia.

Variables	Case (n=76)	Controls (n=152)	P-Value
	Median (IQR)	Median (IQR)	
Age(years)	40(37-48)	40(37-43)	0.155
BMI (kg/m2)	22(21-23.7)	21(20-22)	0.003
RF (+/-)	57/19	0/0	-
TC (mg/dl)	174(162-200)	155(147.5-166.5)	0.000
TG (mg/dl)	145(138-157)	125(115-138)	0.000
HDL-C (mg/dl)	43(40.5-48)	50(47.54)	0.000
LDL-C (mg/dl)	100(93-120)	77(70-86)	0.000
UA (mg/dl)	5.8(5-6.7)	3.8(3.5-4.6)	0.000

5.4. COMPARISON OF ATHEROGENIC INDICES BETWEEN CASE AND CONTROL GROUP

The median and interquartile range values of TC/HDL-C and LDL/HDL-C for RA patients were 3.9(3.5-4.5) and 2.3(2.0-2.7) respectively. The median and interquartile range values of TC/HDL-C and LDL/HDL-C for controls were 3(2.8-3.4) and 1.56 (1.34-1.73), respectively. The calculated atherogenic ratios were statistically significant and higher in cases compared to controls.

Table 4. Comparison of atherogenic indices among case and control group from December 1, 2021 to April 1, 2022 at WCSH, Worabe, Southern Ethiopia.

Variables	Case (n=76)	Controls (n=152	P-Value
	Median (IQR)	Median (IQR)	
TC/HDL-C	3.9(3.5-4.5)	3(2.8-3.4)	0.000
LDL-C/HDL-C	2.3(2.0-2.7)	1.56(1.34-1.73)	0.011

5.5. CORRELATION ANALYSIS OF SERUM LIPID PROFILES WITH URIC ACID IN RA PATIENT

According to spearman coefficient correlation in RA study participants, there was significant moderate positive correlations between UA and TC (ρ=0.692, p<0.001), TG (ρ=0.555, p<0.001) and LDL-C (ρ=0.675, p<0.001). On the other hand, there was Significant moderate negative correlations were found between UA and HDL-C (ρ= -0.449, p<0.001).

Table 5. Bivariate correlation analysis between serum lipid profiles with uric acid in RA patients from December 1, 2021, to April 1, 2022 at WCSH, Worabe, Southern Ethiopia (n=76).

Variables	Bivariate Correlations	
	UA	
TC	P	0.692**
	P	0.000
TG	P	0.555**
	P	0.000
HDL-C	P	-0.449**
	P	0.000
LDL-C	P	0.675**
	P	0.000
TC/HDL-C	P	0.621**
	P	0.000

Note: rho (ρ) = Spearman's rank correlation coefficient, p = p value for correlation, ***** correlation significant at $p \leq 0.05$ level, ****** correlation significant at $p \leq 0.001$ level, TC=total cholesterol, TG=triglycerides, LDL-C = low density lipoprotein-cholesterol, HDL-C=high density lipoprotein-cholesterol, UA=uric acid

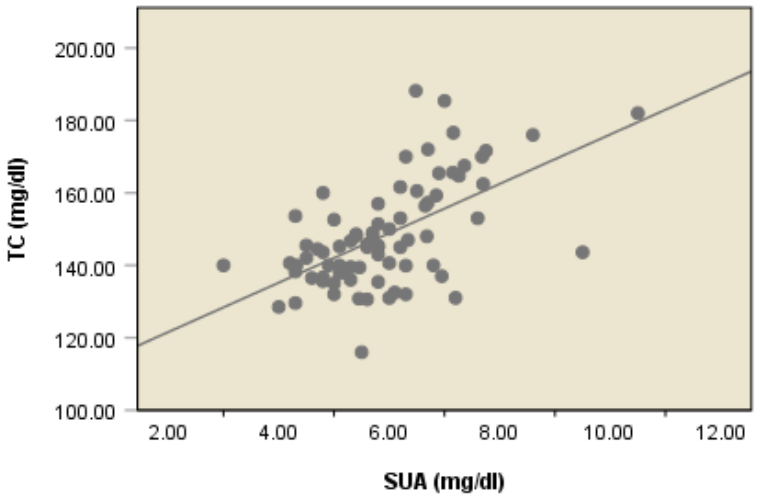

Figure 2a. A scatter plot shows the correlation between TC and SUA values of RA patient

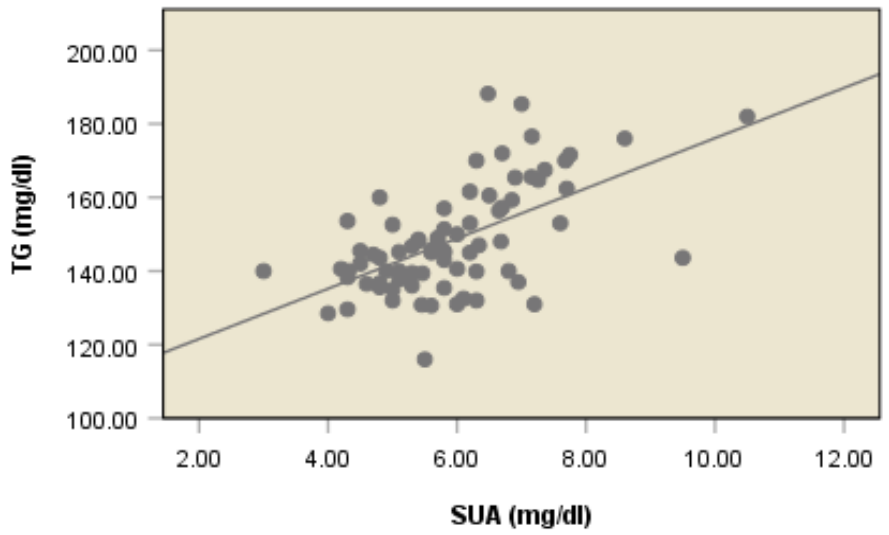

Figure 2b. A scatter plot shows the correlation between TG and SUA values of RA patients

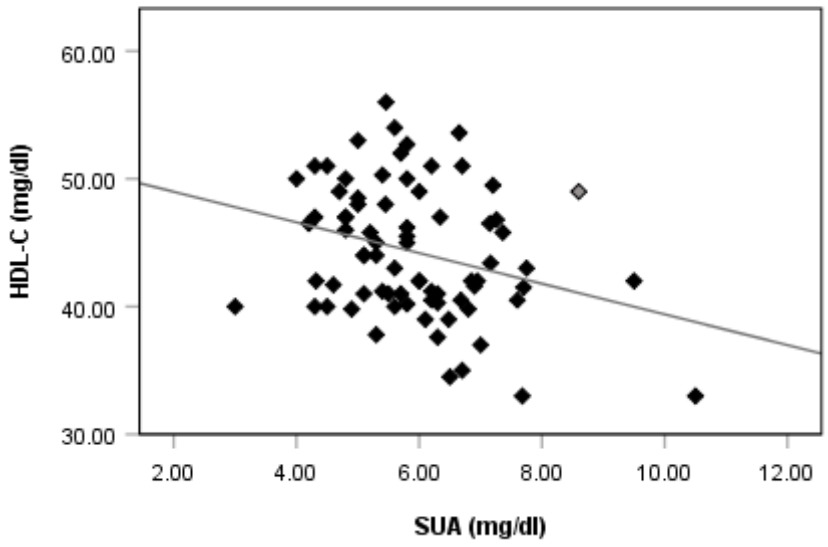

Figure 2c. A scatter plot shows the negative correlation between HDL-C and SUA values of RA patients

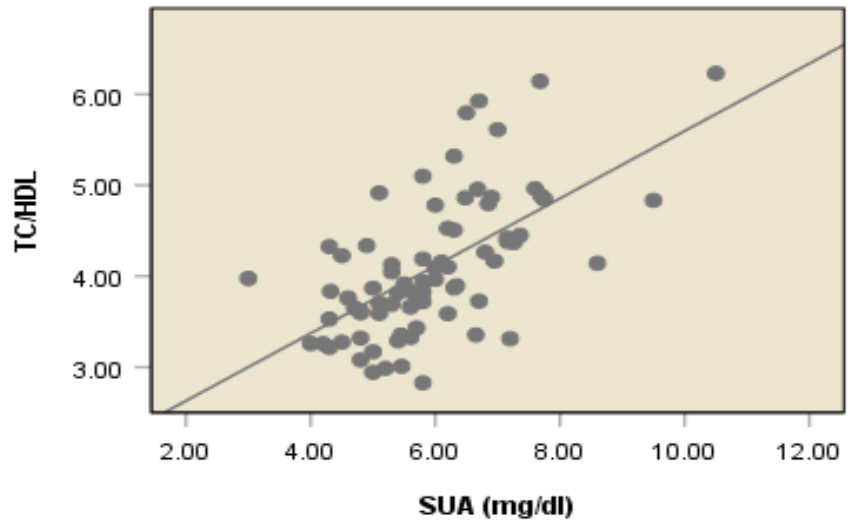

Figure 2d. A scatter plot shows the correlation between TC/HDL-C and SUA values of RA patients

CHAPTER SIX

6. DISCUSSION

Rheumatoid arthritis (RA) is a chronic autoimmune disease affecting the joints. It is characterized by ongoing symmetric inflammation of affected joints, resulting in cartilage destruction, bone erosion, and disability(38). Initially, just a few joints are damaged, but as the condition progresses, more joints are impacted, and extraarticular symptoms are frequently experienced(28). RA is a chronic inflammatory autoimmune disease characterized by inflammation of the synovial tissue of the joints with an unknown etiological factor(53).

In this study, 76 identified rheumatoid arthritis patients were involved. Of the total enrolled RA patients, 80.3% and 19.7% were females and males, respectively, with a female to male ratio of 4.07:1. The age of the patients ranged from 27 to 66 years old, with a median age of 40(37-48) in the study group and 40(40-43) in the control group.

Studies explain that rheumatoid arthritis is an autoimmune disease. According to patients' medical chart, rheumatoid factor test was done for all RA patients. Of the total of 76 rheumatoid arthritis patients, 57 patients were positive for the rheumatoid factor test, whereas 19 patients were negative for the test.

The result of our study shows that the sensitivity of the rheumatoid factor test is 75%. Different types of studies explain the sensitivity and specificity of the rheumatoid factor test for the diagnosis of RA. The sensitivity of the test done by our study agrees with the previous research(54). Emerging data strongly suggests that anti-CCP antibodies (anti-cyclic citrullinated peptide antibodies) have the power to predict the development of RA in patients, but it was not available in our research conducting area.

Lipid levels appear to be changed as a result of RA disease activity. Inflammation may change the constituents of high-density lipoproteins (HDL), reducing their ability to remove cholesterol from atherosclerotic plaque and impairing their anti-oxidant capacity(11). Unfavorable lipid profiles, including higher total cholesterol/HDL-cholesterol ratios or the so-called atherogenic lipoprotein phenotype, characterized by decreased HDL, raised triglycerides, and increased levels of small dense LDL, are associated with RA. Such pro-atherogenic lipid profiles are linked to increased cardiovascular risk beyond LDL-cholesterol levels(12, 13).

This study's major goal was to assess and evaluate the levels of uric acid and serum lipid profiles among RA patients. 76 people with rheumatoid arthritis and 128 apparently healthy people were enrolled in this study as the case and control groups, respectively. According to the study, cases had statistically higher levels of TC, TG, LDL-C, and SUA than controls. In contrast, patients' HDL-C levels were lower than those of controls.

Our study evaluated the dyslipidemia status of both RA patients and control subjects. The study showed that elevation of TC, TG, and LDL-C was seen more frequently among RA patients than controls. In this study, there was a statistically significant lower value of HDLC among RA patients compared to controls. So, the result of our study showed the presence of dyslipidemia among rheumatoid arthritis patients rather than controls, which is similar to studies conducted in Iran, which showed that RA patients are diagnosed with dyslipidemia(50). This is the due to elevation of systemic inflammation among RA patients compared to controls.

In this study, we found that 25 % of RA patients showed higher total cholesterol (≥ 200 mg/dl) values compared to the baseline value of NCETP-ATP III guidelines. An elevated level of TC among cases compared to control was observed in the study done at Tikur Anbessa hospital(47). The possible reasons for the elevation of TC among rheumatoid arthritis patients are chronic inflammation and immune dysregulation.

Of a total of 76 enrolled RA patients, 34.2% showed higher TG compared to the baseline value of the NCETP–ATP III guideline and 5.9% higher as compared to control subjects. This is in line with the study conducted at Tikur Anbessa Specialized Hospital(47).

From the total of 76 enrolled RA patients (15.8%) showed higher LDL-C compared to baseline value of NCETP –ATP III guideline compared to control subjects. Our study showed that the concentration of serum LDL-C was significantly increased compared to controls. But this finding contradicts with the study conducted at Tikur Anbessa Specialized Hospital(47). The possible reason for this variation could be sample size and method of test analysis.

Our results showed a statistically significant ($P< 0.001$) lower value of HDL-c among RA patients compared to controls. A decrease in HDL-C value was seen among rheumatoid arthritis patients compared to controls, agreeing with similar work done in India (49).

In this study, of the total of 76 enrolled RA patients, 19.6% had a lowered HDL-C value. The concentration of HDL-C decreased as inflammatory status increased.

Of the total of 76 enrolled RA patients, 11.8% showed higher SUA compared to control subjects. This fitted with the study done in the United Kingdom(55).

The ratio of TC/HDL-C and LDL/HDL-C was statistically elevated among patients compared to controls. The median and interquartile range of TC/HDL-C among cases and controls was 3.9(3.9-4.5) and 3(2.8-3.4) respectively. The median and interquartile range of LDL/HDL-C among cases and controls were 3(2.8-3.4) and 1.56 (1.34-1.73) (shown in Table 4). This study showed that there was a significant ($P<0.05$) elevation of atherogenic indices among rheumatoid arthritis patients compared to controls. From the total enrolled 76 RA patients, a TC/HDL-C value of 9.8% patients had beyond the baseline value (> 5). The value of TC/HDL-C strongly predicates the presence of future cardiovascular risk than other lipid parameters (56).

In this study, we found significant positive correlations between SUA and TC ($\rho=0.692$), TG ($\rho=0.555$) and LDL-C ($\rho=0.675$), and TC/HDL-C($\rho=0.621$), but negative correlation with HDL-C ($\rho= -0.449$), which is consistence with studies conducted in Bangladesh which showed that SUA was positively correlated with TC (r=0.258), TG (r=0.431), LDL-C (r=0.555) and negatively correlated with HDL-C (r=-0.233)(57). Hyperuricemia is considered to be a mediator of proinflammatory endocrine imbalance in the adipose tissue which may be one of the important factors for dyslipidemia and the inflammatory process that leads to atherogenesis(58).

CHAPTER SEVEN

7. CONCLUSION AND RECOMMENDATION

7.1. CONCLUSION

Rheumatoid arthritis patients had significantly higher levels of TC, TG, LDL-C, TC/HDL-C, LDL-C/HDL-C, and reduced HDL-C values than controls. Thus, it is possible to draw the conclusion that RA patients had dyslipidemia more frequently than controls. In our investigation, we observed that SUA exhibited statistically significant negative correlations with HDL-C but positive correlations with TC, TG, LDL-C, TC/HDL-C, and LDL-C/HDL-C.

7.2. RECOMMENDATION

Based on the above research findings, the following recommendations are forwarded.

- The study should be conducted by measuring additional serological tests other than the rheumatoid factor to easily rule out RA diseases.
- In order to identify cardiovascular risk as early as possible for better patient care, lipid profiles and serum uric acid should be continuously monitored in RA patients.
- A patient with elevated SUA may require screening for renal dysfunction.
- To prevent further health complications, RA patients should focus more on managing their systemic inflammation.

STRENGTH AND LIMITATION OF THE STUDY

STRENGTH OF THE STUDY

Measurement of serum lipid profiles and serum uric acid in rheumatoid arthritis to assess atherosclerosis RA patients hasn't been described in any detail. Therefore, this study is expected to offer the baseline information for further studies on RA in Ethiopia as well as the world.

LIMITATION OF STUDY

- ✓ The study didn't include additional biomarkers like hsCRP and others that are important to investigate cardiovascular risk assessment.
- ✓ Due to feasibility issues, the anti-CCP antibody test was not done.

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ANNEX

ANNEX I: PARTICIPANT INFORMATION SHEET

Title of the Research: Serum lipid profile and uric acid comparison among rheumatoid arthritis patients and apparently health controls at Worabe Comprehensive Specialized Hospital, Worabe, Southern Ethiopia from December 1, 2021, to April 1, 2022

Name of Principal Investigator: Reshad Nuredin

Advisors:

1.Mr. SHIFERAW BEKELE (MSc, Assistant. Prof),

2.Mr. MISGANA BEKELE (MSc),

3.Mr. FANTA OBSA(MSc)

Name of the Organization: Jimma University, Institute of Health, Faculty of Health Sciences, School of Medical Laboratory Sciences.

Introduction: This information sheet is prepared for the aim of explaining the research that you are asked to join with research participant. This information sheet describes about the research.

Aim of the study: To determine the comparison of serum lipid profile and uric acid among rheumatoid arthritis patients and apparently health controls at worabe comprehensive specialized hospital

Procedure: If the patients are agreed to take part in the study, clinical nurse will be given verbal and/or written information about the study and patient will signed on consent form. Patients are kindly requested to give the correct information about them and the necessary measurements will be performed by the assigned nurse. Then 5ml of blood samples will be collected

Risk and discomfort: Participating in this research will not cause more discomfort than is required you could go through for routine examination. If there is any discomfort, we shall offer you necessary medical treatment freely. The amount of blood taken from each volunteer throughout the study period is 5ml which will not affect your health.

Benefits: If you are participating in this study, there may not be direct benefit to you but your participation is likely to help us an important input to find the comparison of serum lipid profile and uric acid among patients with rheumatoid arthritis at worabe comprehensive specialized hospital.

Incentives: There is no financial or material incentive in participating in this study. **Confidentiality**: All information about the patients will be kept confidential. The name of participant will be coded. The information sheet that links the coded number to patient name will be locked inside a computer and it will not be revealed to anyone except your physician and the principal investigator

Participant Rights: You have full right to withdraw from participating in the study at any time before and after consent without explaining the reason and not respond to some or all the questions. Your decision will not affect your right to get health service you are supposed to get otherwise.

Contact Address

If you have any question or concern, you can contact Reshad Nuredin at any time using the following address:

Reshad Nuredin, MSc student at Jimma University, Institute of Health, Faculty of Health Sciences, School of Medical Laboratory Sciences.

Reshad Nuredin

Tel: 09-23-62-24-14 and 09-72-52-70-90

Email: reshadnuran@gmail.com

Jimma, Ethiopia

THANK YOU VERY MUCH!!!

ለጥናቱ ተሳታፊዎች የሚሰጥ መረጃ (AMHARIC VERSION INFORMATION SHEET)
የጥናቱ ርዕስ: በወራቤ አጠቃላይ ስፔሻላይዝድ ሆስፒታል ውስጥ የቁርጥማት ታካሚዎችን የቅባት መጠንና እና የዩሪክ አሲድ ንፅፅር

ተመራማሪ፡ረሻድ ኑረዲን

አማካሪዎች፡

ሸፈረው በቀለ (MSc, Assistant. Prof),

ምስጋና በቀለ (MSc),

ፋንታ አብሳ (MSc)

የተቋሙ ስም፡-ጅማ ዩኒቨርሲቲ፣ ጤና ኢንስቲትዩት ፣ሜዲካል ላቦራቶሪ ትምህርት ክፍል

መግቢያ፡-ይህ የማብራሪያ ቅፅ አሁን እርስዎ እንዲሳተፉ የምንጠይቀዎትን ምርምር ጥናት የሚያብራራ ነው፡፡ በዚህ ጥናት ለመሰሳተፍ ከመወሰንዎ በፊት ይህንን ቅፅ መረጃ ሰብሳቢዎቹ በሚያነቡበት ጊዜ በጥሞና በማድመጥ ጥያቄ ካለዎት በመጠየቅ ትክክለኛዉን መልስ ይመልሱ፡፡ በዚህ ጥናት መሳተፍ ከጀመሩ በኃላ በማንኛውም ጊዜ ጥያቄ ካለዎት መጠየቅ ይችላሉ፡፡

የጥናቱ ዓላማ፡የቅባት መጠንና እና የዩሪክ አሲድ ንፅፅር በቁርጥማት ታካሚዎች ለይ በወራቤ አጠቃላይ ስፔሻላይዝድ ሆስፒታል

የጥናቱ ሂደት፡ይህን ጥናት ለማካሄድ የደም ናሙና በመውሰድ የላብራቶሪ ምርመራ ማደርግ ነው፡፡

ከጥናቱ ጋር የተያያዘ ጉዳት/አለመመቸት፡ እርስዎ በዚህ ጥናት ውስጥ በመሳተፈዎ ለከፋ ጉዳት የሚጋለጡበት ሁኔታ አይኖርም፡፡ደም በሚወሰድበት ወቅት አነስተኛ ህመም ሊሰማወት ይችላል፡፡ እንዲሁም የመቅላት እና የማበጥ ሁኔታ ደም ከተወሰደበት ቦታ ላይ ሊታይ ይችላል፡ ፡ ነገርግን እነዚህ ሁኔታዎች የከፋጉዳት የሚያስከትሉ አይደሉም፡፡

በጥናቱ የመሳተፍ ጥቅም፡ እርስዎ በዚህ ጥናት ላይ በመሳተፍዎ ምንም አይነት የገንዘብ ጥቅም አየገኙም፡፡

የጥናቱ ተሳታፊ ድርሻ፡ በዚህ ጥናት ለመሳተፍ ፍቃደኛ ከሆኑ ከጤናዎ ሁኔታ ጋር የተያያዙ ና ሌሎች የግል መረጃዎችን እንዲሰጡ ይጠየቃሉ፡፡ በመቀጠልም የሰዉነት ክብደተዎን እና የደም ግፊተዎን እንዲለኩና 5 ሚሊ መጠን ያለው የደም ናሙና ለተጠቀሰዉ ዓላማ እንድንወስድ ይጠየቃሉ፡፡

የጥናቱ ተሳታፊዉ መብት፡ በጥናቱ ላይ ለመሳተፍ ባይስማሙ ምንም አይነት ቅጣት የማያስከትል ሲሆን ማንኛውም እርሰዎ ሊያገኙ የሚገባውን ህክምናና ተያያዥ መብት የማያሳጣ መሆኑን እናረጋግጣለን፡፡

የጥናቱ መረጃዎች ምስጥራዊነት፡ እርሰዎን በተመለከተ የምንናገኘውን መረጃ በጥናቱ ወቅትም ሆነ ከዚያ በኃላ ባሉት ጊዜያት እንዲሁም ከጥናቱ የተገኘው መረጃ ሚስጥራዊነት የሚጠበቅ ሲሆን መረጃዎቹም የሚያዙት በስም ሳይሆን በልዩ ኮድ ነው፡፡ይኸው መረጃ በጥንቃቄ የሚያዝ ና የተፈቀደለት ተመራማሪ እና ለህክምና ባለሙያዉ ብቻ ይህም እጅግ አስፈላጊ በሆነ ጊዜ ብቻ ካልሆነ በስተቀር ለሌላ ለማንም ሰው አይሰጥም፡፡ ማንኛውም ከርስዎ ጋር የተያያዘ ውጤት በልዩ ኮድ ብቻ የሚያዝ ሲሆን ውጤቱም ለሳይንሳዊ ዓላማ ብቻ ስም በማይገልፅ ሁኔታ እንዲታተም ይደረጋል፡፡

ስለጥናቱ መረጃ ማግኘት ቢፈልጉ፡ ጥናቱን በተመለከተ ግልጽ ያልሆነ ማንኛዉንም ጥያቄ ካለዎት ነፃ ሆነው ከዚህ በታች ባለው አድራሻ መጠየቅ ይችላሉ፡፡

ረሻድ ኑረዲን

ስልክ: 09-23-62-24-14 ወይም 09-72-52-70-90

ኢሜል: reshadnuran@gmail.com

ጅማ, ኢትዮጵያ

በጣም እናመሰግናለን!!

በመጣሎይ ሊትጋዞን ሰብቸ የስናደ (SILTIGNA VERSION INFORMATION SHEET)

የመጣሎይ መንቄ ዱምከ፡-የቅባትዋ የዩሪክ አሲድ ቂጨ ሃለት በቁርጥማት ነቶ በታንጡ ሰብቻ በወራቤ ኮምፕሬንሲቭ ስፔሽለይዝድ ሁስፒታል፣ ወራቤ፣ ከበ ቶጵየ.

እሞጥላነይ (ሞጣዪ)፡-የኑረዲን ረሻድ

በትጋዞት ዋ ሸገር ባሶት፡- 1. የበቀለ ሸፈረው (MSc, Assistant. Prof),

2. የበቀለ ምስጋና (MSc.),

3. የኡብሳ ፋንታ (MSc.)

የመድረሳይ ሱም፡- የጂመ ዩንቨርስቱ ፈይነት ኢንስቲትዩት ለባራቶሪ አሸር ጋር

መንቄ፡- ሂታይ ዝርዝር ገቦቦ አኩ አቱም የሁን ሌትብሉ ይሳልነመነይ ሱለ ዋ መጣሎ ይዝረዝራነን ግዝ፡፡ ቢታይ መጣሎ ለንበሮት የሁን ተበሎታመ ቀደ ሂነይ ሱለ ሬሬሳ ቲስባስቦናይ ወቅት ፈያኮ በጥነቦት ሱል በለመ በሳሎት ሱታይ ጀዋበ ያቡ፡፡ ቢታይ መጣሎ በጀመሩም ዞፍ በማንም ወቅት ሱል በለመ ተሳሎት ታቀትሉም፡፡

የመጣሎይ ቡር ግዝ ፡-የቅባትዋ የዩሪክ አሲድ ቂጨ ሃለት በቁርጥማት ነቶ በታንጡ ሰብቻ በወራቤ ኮምፕሬንሲቭ ስፔሽለይዝድ ሁስፒታል፣ ወራቤ፣ ከበ ቶጵየ.

የመጣሎይ ሃለት፡ ሂነይ መጣሎ ለሶት ደም በውሰዶት የሊብራቶሪ ምርመራ አሶትን፡፡

ተመጣሎይ በትንዘዛ የማጪያን ደውስ፡ ለመጣሎይ ባሌ ደም ትዮስዱቡም ሀነግን ቲቶቡ ፍርናቸዋ ሀድም ገነ ቁልቁላት አይንበርብም፡፡ ደመ ለውጦት ያጉመ ወክተ ሎክቲክ ኤቲ ጢተ ግዝ ቡሽ የውኖት ሀነግን ሊያብጥ ያቀትላን፡፡ የውን በልዳሌ የፈይነት ሃኪምቸ ሀደም ደውስ ዬለይኮ በሰገርበ ዬውዶን ትዮን፡-አቱም ቢታይ መጣሎ ታንዞታመ ሀነግን ደማም ዋቦ ኡጣተክ ላሎቲ ዬጋየ አለይ፡፡

የመጣሎ ፈይዳ፡ በመጣሎይ በትጋግዞታመ ሀደም ሰንቲበ ኢላትሬክብ፡፡

መጣሎ ሊትገጋዞን ሰብቸ ወጥ፡ ቢታይ መጣሎ ለትገጋዞት የሁን በባመ ተአፊያመ ሃለት የትንዘዙ ዋ ገናም የቢቶ ሬሬሳሶ ቶቦመኮ ይሳሉማን፡፡ ቀጠለኔም የጅስማመ ክባጀ እና ቁኖማመ ትትቃጦም ዋ 5 ሚሊ ቂጪ ያለይ ደም ለጨቀመይ ቡር ገዝ ልሎስዲነ ይሳልነማን፡ :

በሙጣሎይ ሊትገጋዞን ሰብቸ የለይሙይ ሀቅ፡ በሙጣሎይ ለቲገጋዞት በልከሼሙ ሀደም አይነት ቅባጨ አያትኬትለን ትዮን ማነምካ አቱም ሊትረክቡይ እትጌባን ተከሞት ዋ ገነ ገነም ሀቀሙ አያትቁብጣን ሁኖታካ ያትሬግጥና ዬውዲናን፡፡

የሙጣሎይ ሬሬሳሶ ምስጥርነትከ፡ ተቱም በትንዛዘ ይትረከበነይ ሬሬሳ በሙጣሎ ወቅትም ሆነ ታቲም ዞፍ ባለይ ወቅትቸ ሂንኩምንገ ታሙጣሎይ ይትረከባነይ ሬሬሳ ሚስጥርነተከ የትቁረ ትዮን ሬሬሰሶሚ ዬንዙያነይ በሱም ተዮን በሉሌ ኮድን፡፡ሂታሚ ሬሬሳ ፈየኮ ዬንዙያን ና ዋ የትፈቀደኒ ሞጠይ ዋ የፈይነት ሉባም ቢቶን፡፡ ሆነኬም ጥሸት ያትኬሻነን በሆነ ወቅት ግን በልዳሌ ለገነ ለድም ሰብ እሎቡይ፡፡ ማነሚ ተቱም የትንዛዙ ውጣትቸ በሉሌ ኮድ ቢቶ ዬንዙያን ቲዮን ዉጣትምካ ለሳይንሳኜ ገቦቦ ቢቶ ሱም በዬውዲን ሀለት ሊጨቃም ይትራሻን፡፡

ለሙጣሎይ ሬሬሳ ርከቦት ባትኬሼሙ፡ በሱልም የውን በገነ የቲንዛዘ ገቦቦ ጉረ በገባሙ (ዱዱቅ) ለባለቢሙ ለሳሎት በከሼሙ፡-

የኑረዲን ረሻድ በበሎት ተሳሎት ታቀትሎሙ

የስሌክ እልቅ 09-23-62-24-14 ዋ 09-72-52-70-90

ኢሜል: reshadnuran@gmail.com

ጅመ፣ ቶጵየ

ተደሌ ያሾክረማዉ!!

ANNEX- II CONSENT FORM

I confirm that, as I give consent to participate in the study, it is with a clear understanding of the objectives and conditions of the study and with recognition of my right to withdraw from the study if I change my idea. I have been given the necessary information about the research. I have also been assured that I can withdraw my consent at any time without penalty or loss of benefits. The proposal is explained to me in the appropriate language I understand. I_____ do here by give consent to Dr. /Mr. /Mrs. /Miss _____to include me in the proposed research.

Participant code _____

Participant (signature) _____date_____

Name of the data collector_____

Data collector (signature) _____date_____

CONSENT FORM (AMHARIC VERSION)

ከላይ የተፃፈውን የመረጃ ቅፅ አንብቤ የጥናቱ ዓላማና ጥቅም በግልጽ ተረድቻለሁ በማንኛውም ጊዜ ከጥናቱ ያለምንም ችግርና መንገላታት መውጣት እንደምንችል ተገልፆልኛል። ከዚህም በተጨማሪም የጥናቱን ዓላማ በሚገባኝ ቋንቋ ተረድቻለሁ።በዚህ መሠረት ያለጥናት ቡድኑ አባላት ተፅዕኖ በሙሉ ፈቃደኝነት በዚህ ጥናት ውስጥ በመሣተፍ የሚጠበቅብኝን አስተዋፅዎ ለማበርከት በፊርማዬ አረጋግጣለሁ።

የተሳታፊው የሚስጥር ቁጥር ----------

የተሳታፊው ፊርማ ----------ቀን ----------

የመረጃ ሰብሳቢው ስም ----------

የመረጃ ሰብሳቢው ፊርማ ----------ቀን ----------

CONSENT FORM (SILTIGNA VERSION)

ለደር የትከበተይ ሬሬሳ ፈየኮ አነበብኮ የሙጣሎይ ቡር ግዘ (ኡስቤ) ዋ ዴጋያ ፈያኮ ታጀረብኮ በማንም ወቅት ተሙጣሎይ ሀደም ምካት ዋ ጪጥና ሂዮጎት የቃትሊውኮ ተጩቀመኘን። ቲታሚ ገነ የሙጣሎይ ኡሰቤ ቢገባናን አፊ ተጀረብኮ። ቢታሚ አሰነት ያለሀድምከ ግፊት በሙላ በጋጌ ክሶት ቢታይ ሙጣሎ ውስጥ ለቲጋገዞት ዋ ይትቁርብኛነይ ድጋያ ለቦት በመልከቱ ያበይነው።

የትጋገዘይ የቢቶ እልቅ ----------

የትጋገዘይ መልከት ----------አያም ----------

ሬሬሳይ ጪም ያሼይ ሉባም ሱም ----------

ሬሬሳይ ጪም ያሼይ ሉባም መልከት ----------አያም ----------

ANNEX -III PROCEDURES AND PRINCIPLES OF LABORATORY INVESTIGATIONS

TOTAL CHOLESTEROL: Cholesterol was measured enzymatically in serum in a series of coupled reactions that hydrolyzed cholesterol esters and oxidize the 3-OH group of cholesterol. One of reaction products, H2O2 is measured quantitatively in per oxidase catalyzed reaction that produces color. Absorbance was measured at 500nm. The color intensity is proportional to cholesterol concentration. Desirable or normal cholesterol levels was considered to be those below 200mg/dl.

The reaction sequence is as follows.

Cholesterol Easter + H2O —cholesterol Ester Hydrolase→ cholesterol + Fatty acid

Cholesterol + O2—cholesterol oxidase→ cholest-4-en-3-one +H2O2

2H2O2 + 4-amminophenazone —phenol peroxidase→ 4-(pbenzoquinonemonoimino) + 4 H2O

The color intensity of the dye formed is directly proportional to the cholesterol concentration. It is determined by measuring the increase in absorbance.

REAGENTS:

- R1 - PIPES buffer: 225 mmol/L, pH 6.8; Mg2+: 10 mmol/L;
- sodium cholate: 0.6 mmol/L;
- 4-aminophenazone: ≥0.45 mmol/L;
- phenol: ≥12.6 mmol/L;
- fatty alcohol polyglycol ether: 3%;
- cholesterol esterase (Pseudomonas spec.): ≥25 µkat/L (≥1.5 U/mL);
- cholesterol oxidase (E. coli): ≥7.5 µkat/L (≥0.45 U/mL);
- peroxidase (horseradish): ≥12.5 µkat/L (≥0.75 U/mL);
- Diluent NaCl 9%, 50 mL – the diluent cassette is labeled as NACL.

TRIGLYCERIDE: The method was based on the enzymatic hydrolysis of serum triglyceride to glycerol and fatty acids (FFA) by lipoprotein lipase (LPL). The glycerol is phosphorylated by Adenosine triphosphate (ATP) in the presence of glycerol kinase (GK) to form glycerol-3-phosphate (G-3-P) and adenosine diphosphate (ADP). G-3P is then oxidized by glycerophosphate oxidase (GSO) to form dihydroxyacetone phosphate (DHAP) and hydrogen peroxide. A red colored product is formed by peroxidase (POD) catalyzed coupling of 4-amnioantipyrine (4- AA) and phenol with hydrogen peroxide(H2O2), the optical density was read at 540 nm of which is proportional to the concentration of triglyceride in the sample. The reactions as follow

Triglyceride + 3H2O ⎯LPL→Glycerol + 3FFA

Glycerol + ATP ⎯⎯GK→Glycerol-3-p +ADP

Glycerol-3-p +O2 ⎯⎯GPO→ DHAP +H2O2

4-AA + 4phenol ⎯⎯H2O2→ Quinoeimine + H2O

REAGENTS/MATERIALS:

R1 PIPES buffer: 50 mmol/L, pH 6.8; Mg2+: 40 mmol/L; sodium cholate: 0.20 mmol/L; ATP: ≥ 1.4 mmol/L; 4-aminophenazone: ≥ 0.13 mmol/L;4-chlorophenol:4.7 mmol/L; lipoprotein lipase (Pseudomonas spec.); ≥ 83 µkat/L; glycerokinase (Bacillus searothermophilus): ≥ 3 µkat/L; glycerol phosphate oxidase (E. coli): ≥ 41 µkat/L; peroxidase (horseradish): ≥ 1.6 µkat/L preservative.

Procedure: Ten micro liter of serum was mixed in a cuvvate with 1ml of triglyceride mono-reagent R1 then incubated at room temperature for 10 minutes. Then the optical density was read at 540nm against blank and compared with standard triglyceride concentration used as samples.

HIGH DENSITY-LIPOPROTEIN CHOLESTEROL (HDL-C): Homogeneous enzymatic colorimetric test. In the presence of magnesium ions, dextran sulfate selectively forms water-soluble complexes with LDL, VLDL, and chylomicrons which are resistant to PEG-modified enzymes. The cholesterol concentration of HDL-cholesterol is determined enzymatically by

cholesterol esterase and cholesterol oxidase coupled with PEG to the amino groups (approx. 40%). Cholesterol esters are broken down quantitatively into free cholesterol and fatty acids by cholesterol esterase.

In the presence of oxygen, cholesterol was oxidized by cholesterol oxidase to delta 4-cholestenone and hydrogen peroxide.

In the presence of peroxidase, the hydrogen peroxide generated reacts with 4-amino-antipyrine and HSDA to form a purple-blue dye. The color intensity of the blue dye formed is directly proportional to the HDL-cholesterol concentration. It is determined by measuring the increase in absorbance photometrically.

*HSDA = Sodium N-(2-hydroxy-3-sulfopropyl)-3,5-dimethoxyaniline

REAGENTS/MATERIALS:

R1 TAPSOb) buffer; 62.1 mmol/L, pH 7.77; polyanion: 1.25 g/L; EMSE: 1.08 mmol/L; ascorbate oxidase (cucurbita): > 50 µkat/L; peroxidase (horseradish): > 166.7 µkat/L detergent; BSA: 2.0 g/L; preservative.

R2 Bis-Trisc) buffer: 20.1 mmol/L, pH 6.70; cholesterol esterase (microorganism): > 7.5 µkat/L;

cholesterol oxidase (recombinant E. coli): > 7.17 µkat/L; cholesterol oxidase (microorganism): > 76.7 µkat/L; peroxidase (horseradish): > 333 µkat/L; 4-amino-antipyrine: 1.48 mmol/L; BSA 3.0 g/L; detergents; preservative.

b) 2-Hydroxy-N-tris(hydroxymethyl)methyl-3-aminopropanesulfonic acid

c) Bis(2-hydroxyethyl) iminotris(hydroxomethyl)methane

R1 in position B and **R2** in position C

LOW DENSITY LIPOPROTEIN CHOLESTEROL (LDL-C): This automated method for the direct determination of LDL-cholesterol takes advantage of the selective micellary solubilization of LDL-cholesterol by a nonionic detergent and the interaction of a sugar compound and lipoproteins (VLDL and chylomicrons). When a detergent is included in the enzymatic method for cholesterol determination (cholesterol esterase, cholesterol oxidase

coupling reaction), the relative reactivities of cholesterol in the lipoprotein fractions increase in this order: HDL < chylomicrons < VLDL < LDL. In the presence of Mg++, a sugar compound markedly reduces the enzymatic reaction of the cholesterol measurement in VLDL and chylomicrons. The combination of a sugar compound with detergent enables the selective determination of LDL-cholesterol in serum.

Reagents - working solutions

R1 MOPS (3-morpholinopropane sulfonic acid) buffer: 20.1 mmol/L, pH 6.5; HSDA: 0.96 mmol/L; ascorbate oxidase (Eupenicillium spec., recombinant): ≥ 50 µkat/L; peroxidase (horseradish): ≥ 167 µkat/L; preservative

R2 MOPS (3-morpholinopropane sulfonic acid) buffer: 20.1 mmol/L, pH 6.8; MgSO4·7H2O: 8.11 mmol/L; 4-aminoantipyrine: 2.46 mmol/L; cholesterol esterase (Pseudomonas spec.): ≥ 50 µkat/L; cholesterol oxidase (Brevibacterium spec., recombinant): ≥ 33.3 µkat/L; peroxidase

(horseradish): ≥ 334 µkat/L; detergent; preservative

SERUM URIC ACID: determination was based on enzymatic colorimetric test. Uricase cleaves uric acid to form allantoin and hydrogen peroxide. In the presence of peroxidase, 4-aminophenazone is oxidized by hydrogen peroxide to a quinone-diimine dye. The red color intensity of the quinone-diimine formed is directly proportional to the uric acid concentration.

Reagent Preparation (*Reagents are ready to use; no preparation required)

•**R1** reagent. Phosphate buffer: 0.05 mol/L, pH 7.8; TOOS: 7 mmol/L; fatty alcohol polyglycol ether: 4.8 %; ascorbate oxidase (EC 1.10.3.3; zucchini) ≥ 83.5 µkat/L (25 °C); stabilizers

•**R2** reagent. Phosphate buffer: 0.1 mol/L, pH 7.8; potassium hexacyanoferrate (II): 0.3 mmol/L; 4-aminophenazone ≥ 3 mmol/L; uricase (EC 1.7.3.3; Arthrobacter protophormiae) ≥ 83.4 µkat/L (25 °C); peroxidase (POD) (EC 1.11.1.7; horseradish) ≥50 µkat/L (25 °C); stabilizers

ANNEX -IV QUESTIONNAIRE

QUESTIONNAIRE (ENGLISH VERSION)

Code No_____

Part1. Socio-demographic, Anthropometric and clinical information

1. Age ---------- years

2. Sex A. male B. female

3. What is your marital status? A. Single B. Married C. Divorced D. Widowed

4. From which residence you came? A. urban B. Rural

5. What is your educational status? A. Illiterate B. Reading and writing C. Primary school D. Secondary School E. College / University completed

6. Regular physical exercise? A. Yes B. No

7. Height (m) _____

8. Weight (in Kg) _____

9. Body Mass Index (Kg/m2) _____

10. Do you drink Alcohol? A. Yes B. No

11. If yes to the above question, how often did drink alcohol. A Nondrinker B. Non habitual C. Habitual drinker

12. Do you Smoke? A. Yes B. No

13. Do you chew khat? A. Yes B. No

14. Which type of oil is used for your food preparation? A. Palm cooking oil B. Liquid olive oil C. others

15. What is your occupation? A. industry Workers B. House wife C. Farmer D. Merchant

E. Civil servant

Part II. Laboratory investigation result

Test parameter	Result	reference range
TC		
TG		
HDL-C		
LDL-C		
SUA		

QUESTIONNAIRE (AMHARIC VERSION)

የተሳታፊ መላያ ኮድ----------

ክፍል 1፡ የማህበራዊ ና ስነ-ህዝብ ባህሪያት

1. **እድሜዎት ስንት ነው?** _____ **ዓመት**

2. **ጾታ? ሀ. ወንድ ለ. ሴት**

3. **የጋብቻ ሁኔታወስ? ሀ. ያላገባ/ች ለ. ያገባ/ች ሐ. የፈታ/ች መ. የሞተችበት/ የሞተባት**

4. **መኖሪያ ቦታ የት ነው? ሀ. ከተማ ለ. ገጠር**

5. **መደበኛ የትምህርት ደረጃዎ ስንት ነው? ሀ. ማንበብም ሆነ መጻፍ አልችልም ለ. ማንበብ መጻፍ እችላለሁ ሐ. አንደኛ ደረጃ አጠናቅቄያለሁ (1-8) መ. ሁለተኛ ደረጃ አጠናቅቄያለሁ (9-12) ሠ. ኮሌጅ ወይም ዩኒቨርሲቲ አጠናቅቄያለሁ**

6. **መደበኛ የአካል ብቃት እንቅስቃሴ ያድርጋሉ ሀ. አዎ ለ. አላደርግም**

7. **የሰውነት ክብደት ልኬት (ኪ.ግ/ሜ2)** _____

8. **ክብደት (በኪ.. ግ)** _____

9. **ቁመት (በሜትር)** _____

10. **አልኮል ይጠጣሉ ሀ. አዎ ለ. አልጠጣም**

11. **መልሶ አዎ ከሆነ ምን የህል ጊዜ ይጠጣሉ ሀ. አልፎ አልፎ እጠጣለሁ ለ. ሁሌ እጠጣለሁ**

12. **ሲጋራ ያጨሳሉ ሀ. አዎ ለ. አላጨስም**

13.**ጫት ይቅመሉ ሀ. አዎ ለ. አልቅምም**

14. **ምግብ ለማዘጋጀት የሚጠቀሙት የዘይት ዓይነትስ? ሀ. የምረጋ ለ. ፋሳሽ ሐ. ሌላ አይነት**

15. የሚተዳደሩበት የስራ ዓይነት ምንድን ነው; ሀ. ፋብሪካ ሰራተኛ ለ. የቤት እመቤት ሐ. አርሶ አደር መ. ነጋዴ ሠ. የመንግስት ሰራተኛ

ክፍል 2: የላቦራቶሪ ምርመራ ውጤት

የምርመረ አይነት	ውጤት	ሪፈረንስ ሬንጅ
TC		
TG		
HDL-C		
LDL-C		
SUA		

QUESTIONNAIRE (SILTIGNA VERSION)

ሉሌ ኮድ……

ፈስል 1፡ ያመኜ ዋ ስነ-ኡማት ሃለት

1. ኡምር _____ ዘማን

2. ሊገ ሀ. ልጂ ለ. ገረድ

3. የብተር ሃለት ሀ. ብተር አላንዜሆ ለ. ኤንዜሆ ሐ.ተጋፈርኮ መመውት ላዬኛን

4. የንባረት ኤት ሀ. ጌዬን ለ. ከተማን

5. የአሸር ቂጨ ሃለት ሀ. ቅሮት ዋ ክተቦት እለውችል/አልቀራሆ ለ. ቅሮት ዋ ክተቦት ያቀሊው ሐ. 1ኜ መቃም መ. 2ኜ መቃም ሠ.ኮላጅ/ኡንበረስቱ ጃጎ ቀራሆ

6. የጅስመ ተቅላቀሎት ያሶን ሀ. አው ለ. እለዋሽ

7. ቁኖ (ሜ.)----------

8. ክበጀ (ኪ..ግ)----------

9. የጅስም ቂጨ ሀዛት (ኪ..ግ/ሜ^2)----------

10. ሀረቂ ይሰኮን ሀ አው ለ እለውሰች

11. ለደሪ ሱል ጀወባም አው በሆነ ምየህላን ወክተ ትሰኮም ሀ. ስረም እለውሰች ለ. ሀድሀደ ግን ሐ. ሁለም ግን

12. ሱጃረ ይሰኮን ሀ. አው ለ. እለውሰች

13. ጫት ይቂሞን ሀ. አው ለ. እለውቂም

14. አይነካሌን ዘይተ እድገለሎን ሀ ይረጋነይ ለ ፈሳሸይ ሐ ገነ አይነት

15. የመትንዳደሬ ብል ሀ. የፈፍሪከ ብል ለ. የጋር ብል ሐ. እርሰት መ. ዝልዛሎ ሠ. የመንግስትን ብል

ፈስል 2: የላቦራቶሪ ምርመራ ዉጣት

የምርመረ አይነት	**ዉጣት**	**ሪፈረንስ ሬንጅ**
TC		
TG		
HDL-C		
LDL-C		
SUA		

ANNEX- V DECLARATION SHEET

The undersigned declares that this thesis paper complies with the regulations of the University and meets the accepted standards with respect to originality and quality. Principal investigator also agrees to accept responsibility for the scientific ethical and technical conduct of the research project and for provision of required progress reports.

Reshad Nuredin (MSc. candidate) Signature _____ Date_____

Approval of the first Advisor

Mr. Shiferaw Bekele (MSc, Assis. Prof of CLS) Signature _____ Date_____

Approval of the second Advisor

Mr. Misgana Bekele (MSc in CLS) Signature _____ Date _____

Approval of the third Advisor

Mr. Fanta Obsa (MSc in CLS) Signature _____ Date _____

Name of external examiner

MR. ZERIHUN ATARO (Assis. Prof of CLS) Signature _____ Date _____

Name of Internal Assessor

Mr. Waqtola Cheneke (MSc., Asso. Prof. of CLS) Signature _____ Date _____

Head of the School

Signature_____ Date_____