3.1 Materials

3.1.1 Study design:

This was a cross section case control study.

3.1.2 Study area:

The study was conducted in Sudanese diabetic patients in Abdalla Khalil center for diabetes in Khartoum state (Omdurman).

3.1.3 Study period:

The study took a period from March to May 2018.

3.1.4 Ethical consideration:

The study was approved by the Committee of Clinical Chemistry Department at College of Medical Laboratory science of the Sudan University of Science and Technology. A verbal informed consent was obtained from each participant (appendix I).

3.1.5 Study population:

This study included 50 Sudanese diabetic patients, 20 of them were males and the other 30 were females and 45 healthy individuals as control, Age was matched, ranged from (25 to 100) years.

3.1.6 Inclusion Criteria:

Sudanese male and female Who were diagnosed with diabetes mellitus type 2 were enrolled in this study after their approval.

3.1.7 Exclusion Criteria:

Females and males with diabetes mellitus were excluded from this study if they had coronary artery disease, chronic renal disease, heart failure, taking diuretics, taking minerals and multivitamin supplements, pulmonary tuberculosis, patient suffering from cancer or with hepatitis C virus antibody and hepatitis B virus surface antigen, pregnant women, alcohol addiction and smoker were excluded carefully by clinical history.

3.1.8 sampling:

Individuals who voluntarily accepted to participate in the study were enrolled. Data was collected by using questionnaire (Appendix II).

3ml of venous blood was collected from each participants, placed in plane 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.

3.2 Methods:

Estimation of serum GGT, Cholesterol, Triglycerid concentrations using Biosystem analyzer.

3.2.1.GGT Estimation:

Principle of the method:

Gamma-glutamyltransferase (GGT) catalyzes the transfer of gamma-glutamyl group from gamma-glutamyl-3-carboxy-4-nitroanilide to glycylglycine , liberating 3-carboxy-4-nitroaniline . The catalytic concentration is determined from the rate of 3-carboxy-4-nitroaniline formation . (Appendix III).

3.2.2 Cholesterol Estimation:

Principle of method:

Free and esterified cholesterol in the sample originates, by means of coupled reactions described below, a coloured complex that can be measured by spectrophotometry. (Appendix IV).

Cholesterol ester + H2O Chol. esterase Cholesterol + Fatty acid

Cholesterol +1\2 O2 + H2O Chol.oxidase Cholestenone + H2O2

2H2O2 +4-Aminoantipyrine+Phenol peroxidise Quinoneimine+ 4H2O

3.2.3 Triglycerides Estimation:

Principle of method:

Triglycerides in the sample originates, by means of coupled reactions described below, a coloured complex that can be measured by spectrophotometry. (Appendix V).

Triglycerides + H2O lipase Glycerol + Fatty acid

Glycerol + ATP glycerol kinase Glycerol-3-p + ADP

Glycerol-3-p + O2 G-3-P-oxidase Dihidroxyacetone- P + H2O2

2H2O2 + 4-Aminoantipyrine + 4-Chlorophen lipase <u>peroxidase</u> Quinoneimine + 4H2O2

3.3 Quality Control:

Biochemistry Control Serum Level I (cod. 18005, 18009 and 18042) and II (cod. 18007,18010 and 18043) to verify the performance of the measurement procedure. Each laboratory should establish its own internal Quality Control scheme and procedures for corrective action if controls do not recover within the acceptable tolerances.

3.4 Data analysis:

Collected data were analyzed by a computer system using statistical package for social sciences (SPSS) program version 14.