بسم الله الرحمن الرحيم

Shendi University

Faculty of Graduate Studies and Scientific Research

Plasma Chromium level in Sudanese Patients with Diabetes Mellitus

الكروم في المرضى السودانين المصابين بالسكري

A thesis submitted for partial fulfillment for the requirement of M.Sc

Degree Medical Laboratory Sciences in (Clinical Chemistry)

by:

Fatima Awad Alkareem Mohammed Algalal

BSc in Medical Laboratory Sciences , Faculty of Medical Laboratory Sciences , Shendi University

Supervisor:

Dr: Mosab Omer Khalid Mohammed Zeen

Assistant Professor of Clinical Chemistry Faculty of Medical Laboratory Sciences, Shendi University

August 2018

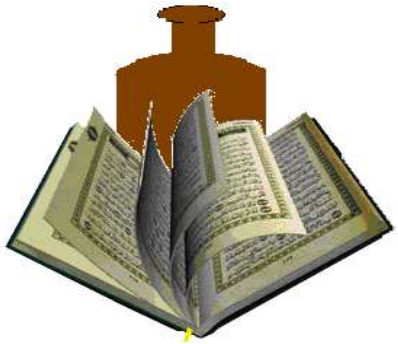
الآية

قال تعالـــــى:

﴿ قُلِ اللَّهُمَّ مَالِكَ الْمُلْكِ تُؤْتِي الْمُلْكَ مَنْ تَشَاءُ وَتَنْزِعُ الْمُلْكَ مِمَّنْ تَشَاءُ وَتُعِزُّ مَنْ تَشَاءُ وَتُذِلُّ مَنْ تَشَاءُ بِيَدِكَ الْخَيْرُ إِنَّكَ عَلَى كُلِّ شَيْءٍ قَدِيرٌ * تُولِجُ اللَّيْلَ فِي النَّهَارِ وَتُولِجُ النَّهَارَ فِي اللَّيْلِ وَتُخْرِجُ الْحَيَّ مِنَ الْمَيِّتِ وَتُخْرِجُ الْمَيِّتَ مِنَ الْحَيِّ وَتَرْزُقُ مَنْ تَشَاءُ بِغَيْرِ حِسَابٍ ﴾

صدق الله العظيم

[آل عمران: 26 - 27].



الإهـــداء

إلى ملهمتي في الحياة...

إلى من كان دعاؤها سر نجاحي

وحنانها بلسم جراحي ...

(أمي الحبيبة)

يامن كلله الله بالهيبة والوقار.. إلى من علمني العطاء بدون انتظار.. إلى من أحمل

إسمه بكل افتخار

(أبي الغالي) الف رحمة و نور عليك

إلى المحبة التي لا تنضب ..

والخير بلا حدود ..

إلى من شاركتهم كل حياتي ..

(أخواني وأخوتي)

الي أروع من جسد الحب بكل معانيه ..

فكان السند والعطاء ..

إلى نصفي الآخر (البراء)

Acknowledgment

First and foremost i would like to thank Allah who gave me the ability to complete this work.

Immeasurable and deepest thank for my supervisor Dr. Mosab Omar for being guide throughout this work and seriously grateful you for your ongoing mentorship, support, and direction.

Special thanks go to my family and everyone who helped me to conduct this study.

Abstract

Diabetes mellitus constitutes one of the most important problems in developing and non-developing countries. There is accumulating evidence that the metabolism of several trace elements is altered in diabetes mellitus, accordingly the study conducted to assess the effect of diabetes mellitus on serum chromium level. **Materials and Methods:** Descriptive cross-sectional study was conducted during the period of March to august 2018. 70 subjects were enrolled in this study; they were classified into 50 subjects as case and 20 as control, serum chromium levels were measured using atomic absorption spectrophotometer.

Results: The mean of serum chromium in diabetic patients when compared with healthy controls observed statistical insignificant difference (p.value 0.106).our results findings there was no effects of duration of Diabetes Mellitus and gender on the serum chromium levels (p.value was 0.541) and (p.value was 0.196). respectivlly.

Conclusion: The mean plasma chromium in diabetic patients when compared with controls found statistical insignificant difference.

Recommendations: Improve health education programs about diabetes and it is complication for population including different age group. Especially to patient with diabetes.

المستخلص

مرض السكري يعتبر من أهم المشاكل الصحية في البلدان النامية وغير النامية . هنالك أدلة أن استقلاب العديد من العناصر الشحيحة بتغير عند الأشخاص المصابين بالسكري . وفقاً لذلك اجريت الدراسة لقياس تركيز الكروم عند مرضى السكري .

المواد والطرق : أجريت دراسة وصفية مستعرضة خلال الفترة من مارس حتى أغسطس 2018م . وشملت الدراسة (70) شخص ، (50) منهم مرضى سكري و (20) شخص أصحاء . وتم قياس تركيز الكروم باستخدام مطيافية الامتصاص الزري .

النتائج : أوضحت الدراسة أن ليس هنالك زيادة إحصائية ذات دلالة في متوسط تركيز الكروم لدى مرضى السكري مقارنة مع الأشخاص الأصحاء القيمة الأحتمالية (0.106) . كما كشفت الدراسة أنه لايوجد تغيير ذو دلالة إحصائيه في متوسط تركيز الكروم مقارنة بالفترة الزمنية لمرض السكري القيم الإحتمالية = (0.541) .

كما بينت الدراسة أنه لايوجد تغيير ذو دلالة إحصائيه في متوسط تركيز الكروم مقارنة بنوع (ذكر ـ أنثى) القيم الإحتمالية = (0.196) .

الخلاصة : خلصت الدراسة إلى أن ليس هناك زياده في متوسط تركيز الكروم عند مرضى السكري .

List of Contents

List of Tables:

List of Figures:

Abbreviations:

ADA.. American Diabetes Association

Co …..Cobalt

Cr …. Chromium

Cu … Copper

CVD ... Cardio Vascular Disease

DM .. Diabetes Mellitus

ECF ….. Extra cellular fluid

Fe … Iron

GAD65 …. Glutamic acid decarboxylase

GDM ….. Gestational diabetes mellitus

HNF1 alpha ….. Human nuclear factor one alpha

HNF4 alpha ….. Human nuclear factor four alpha

IDDM …. Insulin indepent diabetes mellitus

Ifg ….. Impair fasting glucose

Igt…… Impair glucose tolerance

Mg … Magnesium

Mn … Manganese

Mo … Molybdenum

Ni …. Nickel

NIDDM …. Non insulin dependent diabetes mellitus

Pb … Lead

Se … Selemium

SOD … Superoxide dismutase

Zn … Zinc

Chapter One

Introduction

Rational

Objectives

1.1 Introduction:

Diabetes Mellitus is the commonest endocrine disorder encountered in clinical practice. The disease is characterized by distributed metabolism of carbohydrates, lipids and proteins leading to hyperglycemia, ketonemia, acidosis and other metabolic abnormalities and lately the occurrence of other complications, involving small blood vessels, eyes, kidneys, nerves and accelerated atherosclerosis. Diabetes Mellitus may be primary or secondary. Primary diabetes mellitus is generally classified into insulin dependent diabetes mellitus (IDDM) or type 1 Diabetes Mellitus and Non-insulin dependent diabetes mellitus (NIDDM) or type 2 Diabetes Mellitus, Whereas secondary diabetes mellitus may arise from pancreatic disease, stressful situations, over production of hormones, like growth hormone, glucocorticoids, and thyroxin as well drug therapy. Unlike type 1 diabetes mellitus, the pathogenesis of type 2 diabetes mellitus is more complex and less certain. The disease is characterized by a combination of decreased insulin secretion or their actions both of which proceeds and predict the onset of disease. Obesity is a major risk factor for disease. Although environmental factors, both post and prenatal play an important role in determining the risk of disease, a substantial evidence supports that the disease is influenced by inheritance, On molecular basis several uncommon Mandolin forms of type 2 diabetes mellitus have been defined. Type 2 diabetes mellitus, once considered as a rare disease, has been reported to be on rise at an alarming rate, and its prevalence according to WHO estimates that the number of diabetics will be increased over 300 million by year 2025. In Pakistan, population the diabetic patients will be over 11.5 million after a decade if appropriate measures are not taken to control the disease. Insulin, a hypoglycemia factor, requires Chromium as it activates insulin receptor kinase.

It increases insulin sensitivity, glucose utilization and β- cells sensitivity because of its role in insulin activity.(Hajra, Orakzai et al. 2016) .

The term trace elements refer to chemical elements present in a natural material at very small amounts. Chromium is one of the newer essential trace elements. Have a great role in maintaining good health; chromium may have a function in the control of glucose and lipid metabolism. (Shaw 1980) .

Chromium is a transition metal and trivalent; chromium has been referred to as the `glucose tolerance factor' and implicated in the regulation of glucose and lipid metabolism. The exact mechanism of action remains unclear; however, several mechanisms have been proposed to explain the signaling process. Chromium acts as a cofactor or secondary messenger to insulin, improves insulin sensitivity and facilitate glucose utilization by insulin target tissues and chromium also improves insulin affinity to its receptors, and also activates insulin receptor kinases and at the same time inhibiting insulin receptor phosphatases . Chromodulin, the Cr-binding protein, promotes tyrosine kinase activity of insulin receptor during response to insulin.(Ngala, Awe et al. 2018) .

1.2 Rationale:

Diabetes mellitus is common metabolic disease worldwide affecting approx. 150 million people in 2000, and can causes many complications , can affect many subactances in the body such as trace elements.

These study conduct to assess effect of diabetic mellitus in trace element specially chromium, also there is no study perform in shendi locality.

1.3 Objectives:-

1.3.1 General objective:-

- To evaluate plasma chromium level in diabetic patients.

1.3.2 Specific objectives:-

- To measure effect of type of diabetes mellitus on plasma chromium level.
- To measure effect of duration of diabetes mellitus on plasma chromium level.
- To measure effect of gender on plasma chromium level.

Chapter Two

Literature Review

Literature Review

2.1 Carbohydrates:

2.1.1 Classification of carbohydrates:

Carbohydrates are compound containing C, H and O. The general formula of carbohydrate is $C_x(H_2O)_y$. Carbohydrate can be grouped into generic classification based on the number of carbons in the molecule. Another classification of carbohydrate is based on number of sugar units in the chain:

- Monosaccharide's are simple sugars include (glucose and fructose).
- Disaccharides are formed when two monosaccharide units are joined by a glycosidic linkage. Include (maltose, lactose, and sucrose).
- Oligosaccharides and polysaccharides are the chaining of 2 to 10 sugar units, include (starch and glycogen).(Bishop, Fody et al. 2010) .

2.1.2 Chemical properties of carbohydrate:

Some carbohydrates are reducing substances; these carbohydrates can oxidize or reduce other compounds. Carbohydrates can form glycosides' bounds with other carbohydrate and with non-carbohydrates.(Bishop, Fody et al. 2010) .

2.1.3 Glucose metabolism:

Glucose is primary source of energy for human. The nervous system including the brain totally depends on glucose from surrounding extra cellular fluid (ECF) for energy. Nervous tissues cannot concentrate or store carbohydrates; therefore, it is critical to maintain a steady supply of glucose to the tissue. For this reason, the concentration of glucose in the ECF must be maintained in a narrow range. When the concentration falls below a certain level, the nervous tissue loses the primary

energy source and are incapable of maintaining normal function.(Bishop, Fody et al. 2010) .

Carbohydrate metabolism, including glucose metabolism, is regulated by the action and counteraction of the endocrine system. Two hormones, insulin and glucagon, have predominant influence on the pathways of carbohydrate metabolism. The actions of these two hormones counteract each other. Insulin is released in response to increased blood glucose. Insulin acts upon the membrane-bound receptors of tissue cells to allow the movement of glucose in the cell. Insulin also stimulates glycogenesis, lipogenesis, and glycolysis and inhibits glycogenolysis. Glucagon is released in response to a need for increased blood glucose. Glucagon stimulates glycogenolysis and gluconeogenesis. Insulin and glucagons are both produced in the pancreas, insulin in the beta cells and glucagons in alpha cells of the islets of Langerhans. Other hormones also affect carbohydrate metabolism. Epinephrine, a hormone that is released by the adrenal medulla at times of stress, inhibits insulin secretion and stimulates glycogenolysis and lipolysis. Glucocorticoids, such as cortisol, are released from the adrenal cortex to reduce blood glucose concentration by inhibiting gluconeogenesis the absorption of dietary glucose. Thyroxin, a thyroid hormone, increases glycogenolysis and gluconeogenesis and inhibits absorption of dietary glucose through the intestine.(Arneson and Brickell 2007).

2.1.4 Insulin:

Is the primary hormone responsible for the entry of glucose into the cell. It is synthesized by the cells of islets of Langerhans in the pancreas. When these cells detect an increase in body glucose, they release insulin.

The release of insulin causes an increased movement of glucose into the cells and increased glucose metabolism Insulin is normally released when glucose levels are

High and is *not* released when glucose levels are decreased. It decreases plasma glucose levels by increasing the transport entry of glucose in muscle and adipose tissue by way of nonspecific receptors. It also regulates glucose by increasing glycogenesis, lip genesis, and glycolysis and inhibiting glycogenolysis.

Insulin is the only hormone that decreases glucose levels and can be referred to as a hypoglycemic agent.(Bishop, Fody et al. 2010) .

Insulin is stored in the pancreas as the biologically inactive protein proinsulin. Proinsulin is cleaved into the active hormone, insulin, and an inactive peptide, C-peptide.(Arneson and Brickell 2007) .

Insulin, a hypoglycemic factor, requires chromium as it activates insulin receptor kinase. It increase insulin sensitivity, glucose utilization and B cells sensitivity because of its role in insulin activity.(Hajra, Orakzai et al. 2016) .

2.2 Diabetes mellitus:

Diabetes mellitus is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both. The chronic hyperglycemia of diabetes is associated with long-term damage, dysfunction, and failure of various organs, especially the eyes, kidneys, nerves, heart, and blood vessels.(Association 2014) .

Several pathogenic processes are involved in the development of diabetes. These range from autoimmune destruction of the cells of the pancreas with consequent insulin deficiency to abnormalities that result in resistance to insulin action. The basis of the abnormalities in carbohydrate, fat, and protein metabolism in diabetes is deficient action of insulin on target tissues. Deficient insulin action

results from inadequate insulin secretion and/or diminished tissue responses to insulin at one or more points in the complex pathways of hormone action.(Association 2014) .

Impairment of insulin secretion and defects in insulin action frequently coexist in the same patient, and it is often unclear which abnormality, if either alone, is the primary cause of the hyperglycemia. (Association 2014) .

Obesity is a major risk factor for disease, although environmental factors both post and prenatal play an important role in determining the risk of disease, a substantial evidence supports that the disease is influenced by inheritance.(Hajra, Orakzai et al. 2016) Symptoms of marked hyperglycemia include polyuria, polydipsia, weight loss, sometimes with polyphagia, and blurred vision. Impairment of growth and susceptibility to certain infections may also accompany chronic hyperglycemia. Acute, life-threatening consequences of uncontrolled diabetes are hyperglycemia with ketoacidosis or the nonketotic hyperosmolar syndrome.(Association 2014) Long-term complications of diabetes include retinopathy with potential loss of vision; nephropathy leading to renal failure; peripheral neuropathy with risk of foot ulcers, amputations, and Charcot joints; and autonomic neuropathy causing gastrointestinal, genitourinary, and cardiovascular symptoms and sexual dysfunction. Patients with diabetes have an increased incidence of atherosclerotic cardiovascular, peripheral arterial, and cerebrovascular disease.(Association 2014) Hypertension and abnormalities of lipoprotein metabolism are often found in people with diabetes. Type 1 diabetes, the cause is an absolute deficiency of insulin secretion.

Individuals at increased risk of developing this type of diabetes mellitus can often be identified by serological evidence of an autoimmune pathologic process occurring in the pancreatic islets and by genetic markers. In the other, much

more prevalent type 2 diabetes mellitus, the cause is a combination of resistance to insulin action and an inadequate compensatory insulin secretory response. In the latter category, a degree of hyperglycemia sufficient to cause pathologic and functional changes in various target tissues, but without clinical symptoms, may be present for a long period of time before diabetes is detected. During this asymptomatic period, it is possible to demonstrate an abnormality in carbohydrate metabolism by measurement of plasma glucose in the fasting state or after a challenge with an oral glucose load. (Association 2014) .

The degree of hyperglycemia (if any) may change over time, depending on the extent of the underlying disease process. A disease process may be present but may not have progressed far enough to cause hyperglycemia. The same disease process can cause impaired fasting glucose (IFG) and/or impaired glucose tolerance (IGT) without fulfilling the criteria for the diagnosis of diabetes mellitus. In some individuals with diabetes, adequate glycemic control can be achieved with weight reduction, exercise, and/or oral glucose lowering agents. These individuals therefore do not require insulin. Other individuals who have some residual insulin secretion but require exogenous insulin for adequate glycemic control can survive without it. Individuals with extensive cell destruction and therefore no residual insulin secretion require insulin for survival. The severity of the metabolic abnormality can progress, regress, or stay the same. Thus, the degree of hyperglycemia reflects the severity of the underlying metabolic process and its treatment more than the nature of the process itself.(Association 2014) .

2.2.1 Classification of Diabetes Mellitus:

2.2.1.1 Type 1 Diabetes Mellitus:

type 1 diabetes mellitus, insulin-dependent diabetes mellitus (IDDM) (Bishop, Fody et al. 2010) (cell destruction, usually leading to absolute insulin deficiency) Immune-mediated diabetes mellitus. This form of diabetes mellitus, which accounts for only5–10% of those with diabetes mellitus, previously encompassed

by the terms insulin dependent diabetes mellitus, type I diabetes mellitus, or juvenile onset diabetes mellitus, results from a cellular-mediated autoimmune destruction of the cells of the pancreas. Markers of the immune destruction of the cell include islet cell auto antibodies, auto antibodies to insulin, auto antibodies to glutamic acid decarboxylase (GAD65), and auto antibodies to the tyrosine phosphatases IA-2 and IA-2 One and usually more of these auto antibodies are present in 85–90% of individuals when fasting hyperglycemia is initially detected. Also, the disease has strong HLA associations, with linkage to the DQA and DQB genes, and it is influenced by the DRB genes.(Association 2014)

.

These HLA-DR/DQ alleles can be either predisposing or protective. In this form of diabetes mellitus, the rate of cell destruction is quite variable, being rapid in some individuals (mainly infants and children) and slow in others (mainly adults). Some patients, particularly children and adolescents, may present with ketoacidosis as the first manifestation of the disease. Others have modest fasting hyperglycemia that can rapidly change to severe hyperglycemia and/or ketoacidosis in the presence of infection or other stress.(Association 2014) Still others, particularly adults, may retain residual cell function sufficient to prevent ketoacidosis for many years; such individuals eventually become dependent on insulin for survival and are at risk for ketoacidosis. At this latter stage of the disease, there is little or no insulin secretion, as manifested by low or undetectable levels of plasma C-peptide. Immune mediated diabetes mellitus commonly occurs in childhood and adolescence, but it can occur at any age, even in the 8th and 9th decades of life.(Association 2014) .

Autoimmune destruction of cells has multiple genetic predispositions and is also related to environmental factors that are still poorly defined. Although patients are

rarely obese when they present with this type of diabetes mellitus, the presence of obesity is not incompatible with the diagnosis.(Association 2014) .

These patients are also prone to other autoimmune disorders such as Graves' disease, Hashimoto's thyroiditis, Addison's disease, vitiligo, celiac sprue, autoimmune hepatitis, myasthenia gravis, and pernicious anemia.(Association 2014) Type 1 diabetes Mellitus is characterized by lack of insulin production and secretion by the beta cells of the pancreas. One cause of the hyperglycemia of type 1 diabetes mellitus is an autoimmune destruction of the beta cells of the pancreas. The cell mediated response causes infiltration of the pancreas and reduction in the volume of beta cells. As a protein hormone, insulin acts through chemical responses to receptors on the cells of target tissues. In the muscle, insulin stimulates glucose uptake into cells and enhances glycogenesis. In adipose tissue, insulin stimulates glucose uptake into cells and enhances lip genesis. In the liver, insulin has a negative effect, inhibiting gluconeogenesis and glycogenolysis. Auto antibodies are present in the circulation of many individuals with type 1 diabetes mellitus. There appears to be a genetic susceptibility to development of auto antibodies, with certain histocompatibility antigens predominant in the type 1 diabetes mellitus population.(Arneson and Brickell 2007) idiopathic diabetes mellitus is forms of type1 diabetes mellitus have no known etiologies. Some of these patients have permanent insulinopenia and are prone to ketoacidosis, but have no evidence of autoimmunity. Although only a minority of patients with type 1 diabetes mellitus fall into this category, of those who do, most are of African or Asian ancestry. Individuals with this form of diabetes mellitus suffer from episodic ketoacidosis and exhibit varying degrees of insulin deficiency between episodes. This form of diabetes mellitus is strongly inherited, lacks immunological evidence for cell autoimmunity, and is not HLA associated. An absolute

requirement for insulin replacement therapy in affected patients may come and go.(Association 2014) .

2.2.1.2 Type 2 Diabetes Mellitus:

non–insulin-dependent diabetes mellitus (NIDDM)(Bishop, Fody et al. 2010) (ranging from predominantly insulin resistance with relative insulin deficiency to predominantly an insulin secretory defect with insulin resistance) .This form of diabetes mellitus, which accounts for_90–95% of those with diabetes, previously referred to as non-insulin dependent diabetes mellitus, type II diabetes mellitus, or adult-onset diabetes mellitus, encompasses individuals who have insulin resistance and usually have relative (rather than absolute).(Association 2014) insulin deficiency at least initially, and often throughout their lifetime, these individuals do not need insulin treatment to survive. There are probably many different causes of this form of diabetes mellitus. Although the specific etiologies are not known, autoimmune destruction of cells does not occur, and patients do not have any of the other causes of diabetes mellitus.(Association 2014) .

Most patients with this form of diabetes mellitus are obese, and obesity itself causes some degree of insulin resistance. Patients who are not obese by traditional weight criteria may have an increased percentage of body fat distributed predominantly in the abdominal region. Ketoacidosis seldom occurs spontaneously in this type of diabetes; when seen, it usually arises in association with the stress of another illness such as infection. This form of diabetes mellitus frequently goes undiagnosed for many years because the hyperglycemia develops gradually and at earlier stages is often not severe enough for the patient to notice any of the classic symptoms of diabetes mellitus. (Association 2014) .

Nevertheless, such patients are at increased risk of developing macro vascular and micro vascular complications. Whereas patients with this form of diabetes mellitus may have insulin levels that appear normal or elevated, the higher blood glucose levels in these diabetic patients would be expected to result in even higher insulin values had their cell function been normal. Thus, insulin secretion is defective in these patients and insufficient to compensate for insulin resistance.(Association 2014) .

Insulin resistance may improve with weight reduction and/or pharmacological treatment of hyperglycemia but is seldom restored to normal The risk of developing

this form of diabetes mellitus increases with age, obesity, and lack of physical activity. It occurs more frequently in women with prior GDM and in individuals with hypertension or dyslipidemia, and its frequency varies in different racial/ ethnic subgroups. It is often associated with a strong genetic predisposition, more so than is the autoimmune form of type 1 diabetes mellitus. However, the genetics of this form of diabetes mellitus are complex and not clearly defined.(Association 2014) .

Type 2 diabetes mellitus is characterized by decline in insulin action due to the resistance of tissue cells to the action of insulin. The problem is intensified by the inability of the beta cells of the pancreas to produce enough insulin to counteract the resistance. Thus, type 2 diabetes mellitus is a disorder of both insulin resistance and relative deficiency of insulin.(Arneson and Brickell 2007) .

2.2.1.3 Gestational Diabetes Mellitus (GDM):

GDM is defined as any degree of glucose intolerance with onset or first recognition during pregnancy. The definition applies regardless of whether insulin

or only diet modification is used for treatment or whether the condition persists after pregnancy. It does not exclude the possibility that unrecognized glucose intolerance may have antedated or begun concomitantly with the pregnancy. GDM complicates _4% of all pregnancies in the U.S., resulting in 135,000 cases annually. The prevalence may range from 1 to 14% of pregnancies, depending on the population studied. GDM represents nearly 90% of all pregnancies complicated by diabetes. Deterioration of glucose tolerance occurs normally during pregnancy, particularly in the 3rd trimester.(Association 2014) .

Gestational diabetes mellitus is similar in etiology to type 2 diabetes mellitus; however, it is defined as diabetes mellitus that is diagnosed in pregnancy. Pregnancy is associated with increased tissue cell resistance to insulin. Most pregnant women will compensate with increased secretion of insulin; those individuals who are unable to compensate may develop gestational diabetes mellitus. The hyperglycemia of gestational diabetes mellitus diminishes after delivery; however, the individual who has developed gestational diabetes mellitus is at higher risk for the development of type 2 diabetes mellitus thereafter.(Arneson and Brickell 2007) .

2.2.1.4 Genetic defects of beta cell function:

Several form of diabetic mellitus state may be associated with monogenetic defects in beta cell function, frequently characterized by onset of mild hyperglycemia at an early age (generally before age 25 years). They are usually inherited in an autosomal dominant pattern. Patients with these forms of diabetes mellitus, formerly referred to as maturity onset diabetes mellitus of the young (MODY), have impaired insulin secretion with minimal or no defect in insulin action.

Abnormalities at three genetic loci on different chromosome have now been characterized. The most common form is associated with mutation on chromosome 12 in a hepatic nuclear transcription factor referred to as HNF1alpha (54). A second form associated with mutation in the glucokinase gene on chromosome 7p (55,56). Glucokinase converts glucose to glucose 6_phosphate, the metabolism of which in turn stimulates insulin secretion by the beta cell. Thus, glucokinase serves as the "glucose sensor" for the beta cell. Because of defects in glucokinase gene, increase levels of glucose are necessary to elicit normal levels of insulin secretion. A third form is associated with mutation in the HNF4 alpha gene on chromosome 20q(57).(Organization 1999) .

2.2.2 Complications of Diabetes Mellitus:-

Vascular disease is a common complication of diabetes mellitus. Macro vascular disease due to abnormalities of large vessels may present as coronary artery, cerebrovascular or peripheral vascular insufficiency. The condition is probably related to alterations in lipid metabolism and associated hypertension. The most common cause of death is cardiovascular disease, including myocardial infarction.(Crook 2012) .

2.2.2.1 Acute Complications:-

Acute complications include diabetic ketoacidosis, hyperosmolar hyperglycemic nonketotic coma, and hypoglycemia.(Loewen and Haas 1991) .

2.2.2.2 Chronic Complications:-

The chronic complications of diabetes mellitus due to damage in small blood vessels include damage to the eyes, kidneys, and nerves. Damage to the eyes, known as diabetic retinopathy, is caused by damage to the blood vessels in the retina of the eye, and can result in gradual vision loss and blindness. Damage to the kidneys, known as diabetic nephropathy, can lead to tissue scarring, urine

protein loss, and eventually chronic kidney disease, sometimes requiring dialysis or kidney transplant. Damage to the nerves of the body, known as diabetic neuropathy, is the most common complication of diabetes mellitus, The symptoms can include numbness, tingling, pain, and altered pain sensation, which can lead to damage to the skin. Diabetes-related foot problems (such as diabetic foot ulcers) may occur, and can be difficult to treat, occasionally requiring amputation.(Rother 2007)and macrovascular Disease, medium to large size vessels become blocked (atherosclerosis) or vessel walls thicken (arteriosclerosis) reducing or blocking the flow of blood. These vessels supply blood to the legs (peripheral vascular disease), brain (cerebral vascular disease) heart (coronary vascular disease). Risk of heart disease may also be greater in Type 1 diabetics. Several risk factors, obesity, cholesterol level, inactivity, hyperinsulinemia combined with diabetes mellitus put an individual more at risk for developing cardiovascular disease.(Ozougwu, Obimba et al. 2013) .

2.2.2.3 Micro vascular Complications of Diabetes Mellitus:

2.2.2.3.1 Diabetic retinopathy:

Diabetic retinopathy may be the most common micro vascular complication of diabetes. It is responsible for ~ 10,000 new cases of blindness every year in the United States alone.1 The risk of developing diabetic retinopathy or other microvascular complications of diabetes mellitus depends on both the duration and the severity of hyperglycemia. Development of diabetic retinopathy in patients with type 2 diabetes mellitus was found to be related to both severity of hyperglycemia and presence of hypertension in the U.K, retinopathy includes such features as small hemorrhages in the middle layers of the retina. They clinically appear as “dots” and therefore are frequently referred to as “dot hemorrhages.” Hard exudates are caused by lipid deposition that typically occurs at the margins of hemorrhages. (Fowler 2008) .

2.2.2.3.2 Diabetic nephropathy:

Diabetic nephropathy is the leading cause of renal failure in the United States. It is defined by proteinuria > 500 mg in 24 hours in the setting of diabetes mellitus, but this is preceded by lower degrees of proteinuria, or "micro albuminuria." Micro albuminuria is defined as albumin excretion of 30–299 mg/24 hours. Without intervention, diabetic patients with micro albuminuria typically progress to proteinuria and overt diabetic nephropathy. This progression occurs in both type

1 and type 2 diabetes mellitus. The pathological changes to the kidney include increased glomerular basement membrane thickness, micro aneurysm formation, meningeal nodule formation (Kimmelsteil-Wilson bodies), and other changes.(Fowler 2008) .

2.2.2.3.3 Diabetic neuropathy

Diabetic neuropathy is recognized by the American Diabetes Association (ADA) as "the presence of symptoms and/or signs of peripheral nerve dysfunction in

People with diabetes mellitus after the exclusion of other causes. As with other microvascular complications, risk of developing diabetic neuropathy is proportional to both the magnitude and duration of hyperglycemia, and some individuals may possess genetic attributes that affect their predisposition to developing such complications.(Fowler 2008) .

2.2.2.4 Macro vascular Complications of Diabetes:

The central pathological mechanism in macro vascular disease is the process of

Atherosclerosis, which leads to narrowing of arterial walls throughout the body. Atherosclerosis is thought to result from chronic inflammation and injury to the arterial wall in the peripheral or coronary vascular system. In response to endothelial injury and inflammation, oxidized lipids from lower density lipoprotein particles accumulate in the endothelial wall of arteries. In addition to

atheroma formation, there is strong evidence of increased platelet adhesion and hypercoagulability in type 2 diabetes mellitus. Impaired nitric oxide generation and increased free radical formation in platelets, as well as altered calcium regulation, may promote platelet aggregation. Diabetes mellitus increases the risk that an individual will develop cardiovascular disease (CVD). Type 2 diabetes mellitus typically occurs in the setting of the metabolic syndrome, which also includes abdominal obesity, hypertension, hyperlipidemia, and increased coagulability. These other factors can also act to promote CVD.(Fowler 2008) .

2.3 Trace elements:

The human body is composed of elements which can be roughly divided into abundant elements and trace elements. Abundant elements consist of the major elements that are involved in the formation of covalent bonds and are important constituents of tissues(oxygen, carbon, hydrogen, nitrogen, etc.), and semi-major elements, which often exist in the ionic state, and are involved in functions of the living body through maintenance of osmotic pressure and membrane potentials (potassium, sodium, etc.). Major elements account for 96% of the total body weight, and the semi-major elements account for 3 to 4% of the total body weight. Deficiency of major elements can lead to nutritional disorders, and their presence in excess can cause obesity. Deficiencies or excess states of semi-major elements often result in water and electrolyte abnormalities. Essential trace elements of the human body include zinc (*Zn*), copper (*Cu*), selenium (*Se*), chromium (*Cr*), cobalt (*Co*), iodine (*I*), manganese (*Mn*), and molybdenum (*Mo*). Although these elements account for only 0.02% of the total body weight, they play significant roles, e.g., as active centers of enzymes or as trace bioactive substances. A major outcome of trace element deficiencies is reduced activity of the concerned enzymes.(Wada 2004) .

The term trace elements refer to chemical elements present in a natural material at very small amounts. In analytical chemistry, a trace element is an element in a sample that has an average concentration of <100 parts per million (ppm) measured in atomic count or <100 μg/g. In biochemistry, a trace element is a dietary mineral that is needed in very minute quantities for the proper growth, development, and physiology of the organism.(Al-Fartusie and Mohssan 2017) Trace elements have several important roles in human bodies, some are essential for enzymes reactions where they attract and facilitate conversion of substrate molecules to specific end products. Moreover, some of them donate or accept electrons in redox reactions that are of primary importance in the generation and utilization of metabolic energy. Some of them have structural roles and responsible for the stability of important biological molecules. Furthermore, some trace elements have important actions throughout biological processes, for example, ıron (Fe) which can bind, transport, and release oxygen in the body [2,3]. In fact, although the trace elements are essential components of biological activities, the excessive levels of these elements can be toxic for the body health and may lead to many fatal diseases, such as cancers. In this review article, we will describe the properties and biological important of a variety of trace elements.(Al-Fartusie and Mohssan 2017) .

2.3.1 Chromium *(Cr)*:

Chromium is a transition metal and trivalent ,chromium has been referred to as the `glucose tolerance factor' and implicated in the regulation of glucose and lipid metabolism (Ngala, Awe et al. 2018) Chromium is an insulin cofactor. Chromium deficiency can occur on long-term parenteral nutrition, leading to glucose intolerance and neuropathy.(Crook 2012) .

Cr is a chemical element with symbol Cr and atomic number 24. It is a steely-gray, lustrous, hard, and brittle metal. It has an atomic weight of 52.0. In 1797, chromium oxide was discovered by the French pharmacist and chemist Louis Nicolas Vauquelin. In the following year 1798, Vauquelin discovered that he could isolate metallic chromium by heating the oxide in a charcoal oven, which making him the discoverer of the element.(Al-Fartusie and Mohssan 2017) .

Cr is a trace element that humans require in trace amounts. It is found primarily in two forms: Trivalent (chromium III), which is biologically active and found in food and hexavalent (chromium VI), a toxic form that results from industrial pollution. In 2001, Dietary Reference Intakes for chromium were established. Adequate intakes of chromium are 35 mg/day for adult males and 25 mg/day for adult females. Cr is widely distributed in the food supply, but most foods provide only small amounts of it. It is found in egg yolks, whole-grain products, high-bran breakfast cereals, coffee, nuts, green beans, broccoli, meat, and brewer's yeast. (Al-Fartusie and Mohssan 2017) .

Cr levels in biological matter have been studied extensively. It has been found that chromium produces significant increases in enzyme activity and serves an important function in carbohydrate metabolism, stimulation of fatty acid and cholesterol synthesis from acetate in the liver, and improved sugar metabolism through the activation of insulin. In addition, it has been found that chromium renders the body's tissues more sensitive to insulin. It is a critical cofactor in the action of insulin. In fact, the actual chromium deficiency in humans is rare. Despite that, some studies reported that chromium deficiency is associated with glucose intolerance and insulin resistance in patients on long-term parenteral nutrition. Furthermore, it has been reported that chromium deficiency may be the reason to an increase in hematological parameters (hemoglobin, hematocrit, erythrocytes, leukocytes, and mean erythrocyte volume).(Al-Fartusie and Mohssan 2017) .

2.3.1.1 Chromium and blood sugar:

Trivalent chromium enhances the effects of insulin. The body secretes the hormone

Insulin in response to rising levels of blood sugar. Insulin binds to receptors on cell membranes thus stimulating the cells to take in more glucose (blood sugar). This not only clears excess insulin from the blood , but also assists the cells in obtaining blood sugar. Compounds that assist insulin in clearing glucose from the blood are called glucose tolerance factors. Some glucose tolerance factors contain chromium. Trivalent chromium also enhances the ability of insulin to remove fats from the

blood .(Blake 2008) Trivalent chromium is known to enhance insulin action and is necessary for optimal carbohydrate and lipid metabolism.(Lipko and Debski 2018)

A decreased response to insulin can result in impaired glucose tolerance . A more

serious insulin resistance is known as type 2 diabetes mellitus. The clinical signs of type 2 diabetes mellitus include elevated blood sugar levels and insulin resistance.

Cell membranes have insulin receptors . The sensitivity of this receptor to insulin can be improved by chromium. Insulin binds to the insulin receptor in the cell membrane to activate the receptor . The activation of the insulin receptor enables glucose and chromium to enter the cell . Chromium binds to the insulin receptor and enhances its activity. With available chromium, more glucose enters the cell. This is how chromium availability helps to relieve impaired glucose tolerance and type 2 diabetes.(Blake 2008) .

2.3.1.2 Chromium deficiency:

Chromium deficiency is difficult to determine because of the lack of accurate tests

for chromium status. Chromium deficiency may be a contributing factor in both type 2 diabetes and impaired glucose tolerance. Heavy exercise may increase the amount of chromium needed.(Blake 2008) .

2.3.1.3 Food sources for chromium:

Data on the chromium content of foods is limited. Furthermore, the amount of Chromium in foods is quite variable. Good sources of chromium include whole Grain products, broccoli, green beans, grape juice, and spices. Processed foods And foods high in sugar are poor sources of chromium. Foods high in sucrose And fructose increase chromium loss. Vitamin C in amounts of 100 mg or more increase the absorption of chromium.(Blake 2008) .

2.3.1.4 Toxicity of chromium:

As noted above, hexavalent chromium is known to cause cancer and skin irritation. There have been no convincing reports of toxicity from the trivalent chromium in food or supplements. Therefore, the Food and Nutrition Board has not set an upper level for chromium. Several studies have shown a lack of side effects from supplementation of 1000 mcg for several months. People with kidney or liver disease should be cautioned to take lower doses. Normal supplementation levels are between 60 mcg and 120 mcg of chromium. Chromium is a valuable trace mineral because it boosts the power of insulin.(Blake 2008) .

2.3.1.5 Adequate intake male's females for chromium: for all age (mcg/day)

- Infants 0–12 months (0.2_5.5)
- Children 1–13 years (11_ 25)
- Adults (20_35)
- Pregnancy (30)

2.3.2 Zinc *(Zn)*

Zn is an essential trace element that functions as a cofactor for certain enzymes involved in metabolism and cell growth, it is found in nearly 300 specific enzymes. As a component of many enzymes, Zn is involved in the metabolism of proteins, carbohydrates, lipids, and energy. Zn is vital for the healthy working

of many of the body's systems; it plays an essential role in numerous biochemical pathways. It is particularly important for healthy skin and is essential for a healthy immune system and resistance to infection. Zn plays a crucial role in growth and cell division where it is required for protein and DNA synthesis, in insulin activity, in the metabolism of the ovaries and testes, and in liver function.(Al-Fartusie and Mohssan 2017) .

Zinc is an important component of superoxide dismutase (SOD). Superoxide dismutase is an important antioxidant enzyme that converts superoxide free radicals into oxygen and hydrogen peroxide. Hydrogen peroxide is less dangerous as a free radical and can be further degraded into water and oxygen. (Blake 2008).

2.3.3 Selenium *(Se)*

Se is a vital trace element for human body health, where it is found at the active site of a wide range of selenoproteins as selenocysteine. It is an important component of the antioxidant enzymes such as glutathione peroxides and thioredoxin reeducates. Although selenium deficiency is rare in healthy human, it is a very toxic if taken in excess amounts. It has been established that dietary selenium is important for a healthy immune system, where it enhances T-lymphocyte immune responses. It has been found that there is a relationship between low blood levels of Se and increased cardiovascular disease mortality. Furthermore, it has been reported that the lack of selenium is the main reason of Keshan disease. On other hand, there is strong evidence that Se has a protective effect against some forms of cancer such as colon, prostate, and breast.(Al-Fartusie and Mohssan 2017) .

Selenium is used in the body in antioxidant enzymes, by the immune system,

And in conjunction with iodine to control metabolism. needed to convert thyroxin to the more active thyroid hormone. (Blake 2008) .

2.3.4 Copper *(Cu)*

Cu is an essential constituent of several enzymes such as cytochrome oxidase, monoamine oxidase, catalase, peroxidase, ascorbic acid oxidase, lactase, tyrosines, and superoxide dismutase (SOD). Moreover, due to its presence in a

wide variety of enzymes, Cu is involved in many metabolic reactions. For example, the presence of Cu in the SOD helps in the conversion of superoxide to oxygen and hydrogen peroxide. Cu is an essential micronutrient necessary for the hematologic and neurologic systems. It is necessary for the growth and formation of bone, formation of myelin sheaths in the nervous systems, helps in the incorporation of Fe in hemoglobin, assists in the absorption of Fe from the gastrointestinal tract, and in the transfer of Fe from tissues to the plasma. (Al-Fartusie and Mohssan 2017) Copper needed for energy production, collagen synthesis, iron transport, and as an antioxidant. (Blake 2008) .

Other trace minerals may be needed in tiny amounts. However, in large amounts, they can be toxic. these elements like ***(Magnesium (Mg), Manganese (Mn), Nickel (Ni), Cobalt (Co), Lead (Pb),*** **molybdenum , iodine , fluorine , silicon**).

2.4 Previous studies:

2.4.1 Concentrations of seven trace elements in different hematological matrices in patients with type 2 diabetes as compared to healthy controls

This study aimed to compare the trace element status of patients with type 2 diabetes (n=53) with those of non-diabetic healthy controls (n=50). The concentrations of seven trace elements were determined in the whole blood, blood plasma, erythrocytes, and lymphocytes of the study subjects. Vanadium and iron levels in lymphocytes were significantly higher in diabetic patients as compared to controls (p<0.05 for iron and p<0.01 for vanadium). In contrast, lower manganese (p<0.01) and selenium (p<0.01) concentrations were detected in lymphocytes derived from patients with type 2 diabetes versus healthy subjects. Furthermore, significantly lower chromium levels (p<0.05) were found in the plasma of diabetic individuals as compared to controls. Trace element concentrations were not dependent on the degree of glucose control as determined by correlation analysis between HBA_{1c} versus metal levels in

the four blood fractions. In summary, this study primarily demonstrated that trace element levels in lymphocytes of patients with type 2diabetes could deviate significantly from controls, whereas, in general, no considerable differences could be found when comparing the other fractions between both patient groups. Therefore, it seems reasonable to analyze metal levels in leukocytes to determine trace element status in patients with type 2 diabetes and perhaps in other diseases.(Ekmekcioglu, Prohaska et al. 2001) .

2.4.2 Copper, Chromium, Manganese, Iron, Nickel, and Zinc Levels in Biological Samples of Diabetes Mellitus Patients

There is accumulating evidence that the metabolism of several trace elements is altered in diabetes mellitus and that these nutrients might have specific roles in the pathogenesis and progress of this disease. The aim of present study was to compare the level of essential trace elements, chromium (Cr), copper (Cu), iron (Fe), manganese (Mn), nickel (Ni), and zinc (Zn) in biological samples (whole blood, urine, and scalp hair) of patients who have diabetes mellitus type 2 ($n = 257$), with those of non-diabetic control subjects ($n = 166$), age ranged (45–75) of both genders. The element concentrations were measured by means of an atomic absorption spectrophotometer after microwave-induced acid digestion. The validity and accuracy was checked by conventional wet-acid-digestion method and using certified reference materials. The overall recoveries of all elements were found in the range of (97.60–99.49%) of certified values. The results of this study showed that the mean values of Zn, Mn, and Cr were significantly reduced in blood and scalp-hair samples of diabetic patients as compared to control subjects of both genders ($p < 0.001$). The urinary levels of these elements were found to be higher in the diabetic patients than in the age-matched healthy controls. In contrast, high mean values of Cu and Fe were

detected in scalp hair and blood from patients versus the non-diabetic subjects, but the differences found in blood samples was not significant ($p < 0.05$). These results are consistent with those obtained in other studies, confirming that deficiency and efficiency of some essential trace metals may play a role in the development of diabetes mellitus.(Kazi, Afridi et al. 2008) .

2.4.3 Zinc, Copper, Iron, and Chromium Concentrations in Young Patients with Type 2 Diabetes Mellitus

Homeostasis of trace elements can be disrupted by diabetes mellitus. On the other hand, disturbance in trace element status in diabetes mellitus may contribute to the insulin resistance and development of diabetic complications. The aim of present study was to compare the concentration of essential trace elements, zinc, copper, iron, and chromium in serum of patients who have type 2 diabetes mellitus ($n = 20$) with those of non-diabetic control subjects ($n = 20$). The serum concentrations of zinc, copper, iron, and chromium were measured by means of an atomic absorption spectrophotometer (Shimadzu AA 670, Kyoto, Japan) after acid digestion. The results of this study showed that the mean values of zinc, copper, and chromium were significantly lower in the serum of patients with diabetes as compared to the control subjects ($P < 0.05$). Our results show that deficiency of some essential trace elements may play a role in the development of diabetes mellitus.(Basaki, Saeb et al. 2012) .

Chapter Three

Material and Methods

Materials and Method

3.1 Study design:

A cross-sectional descriptive study, conducted during the period between March and May 2018.

3.2 Study area:

The study was conducted in Shendi locality, which is located in the river Niles state; it is about 150 kilometers from Khartoum.

3.3 Data collection tools:

The information's about patients diagnosed as diabetes obtained from a questionnaire, involved some questions including (age, sex, type of diabetic and duration of disease ....etc.).

3.4 population sampling:

A fifty venous blood sample were obtained from known Diabetic patient as case group and 20 venous blood samples were obtained from healthy individual as control group.

3.5 Exclusion criteria:

We exclude patients with renal failure and patients treated with vitamins.

3.6 Material and instrument:

- Cotton.
- Syringe.
- Heparinized containers.
- Plain containers.
- Automatic pipettes.
- Centrifuge.

3.7 atomic absorption spectrophotometer:

Is a spectroanalytical procedure for the quantitative determination of chemical elements using the absorption of optical radiation (light) by free atoms in the gaseous state. In analytical chemistry the technique is used for determining the concentration of a particular element (the analyte) in a sample to be analyzed. AAS can be used to determine over 70 different elements in solution, or directly in solid samples via electro thermal vaporization, and is used in pharmacology, biophysics and toxicology research.

3.8 Principle of atomic absorption spectrophotometer:

Brief According to manufacture, electron of the atom promoted to higher orbital (excited state) for a short period of time by absorbing light energy of specific wavelength, as number of atoms in light path increases the amount of light absorbed also increases, By measuring the amount of light absorbed a quantitative determination of the amount of analyte can be made.

3.9 Sample collection and preparation:

A volume of 3 ml of venous blood sample was collected from all patients in lithium heparin containers. Using a disposable syringe and aseptic standard non-traumatic vein puncture technique was applied, blood was emptied into a sterile containers. And then centrifuged at 4000 rpm for 10 minutes, then the plasma was separated and transferred into a new container and processed. Plasma was stored until analysis.

3.10 Analysis of chromium:

Plasma chromium concentration were determined using flame atomic absorption spectrophotometer (buck scientific 210\211 VGP atomic absorption _England) .

Plasma samples were diluted with deionized water. The analysis was performed against control sample, standard prepared in glycerol to approximate the viscosity characteristics of diluted samples. For determination of plasma chromium, samples were diluted 1\5 with deionized water.

3.11 Data analysis: -

The data were compared by using statistical analysis performed with Statistical Package for Social sciences (SPSS-s) software version 20 to compare means and standard deviation of chromium.

3.12 Ethical considerations:-

Procedure of blood sampling was explained to the participants. All participants were informed about the research objectives and procedures during the interview period. Verbal informed consent was obtained from all participants.

Chapter Four

Results

4. Results

Table (4 – 1): the mean and Standard deviation of plasma chromium level in case and control group:-

	Frequency	Mean (mg\dl)	Std	p. value
Test	50	0.0948	0.03699	0.106
Control	20	0.0785	0.03911	

- T-test was used for analysis.
- P. value > 0.05 is considered insignificant.

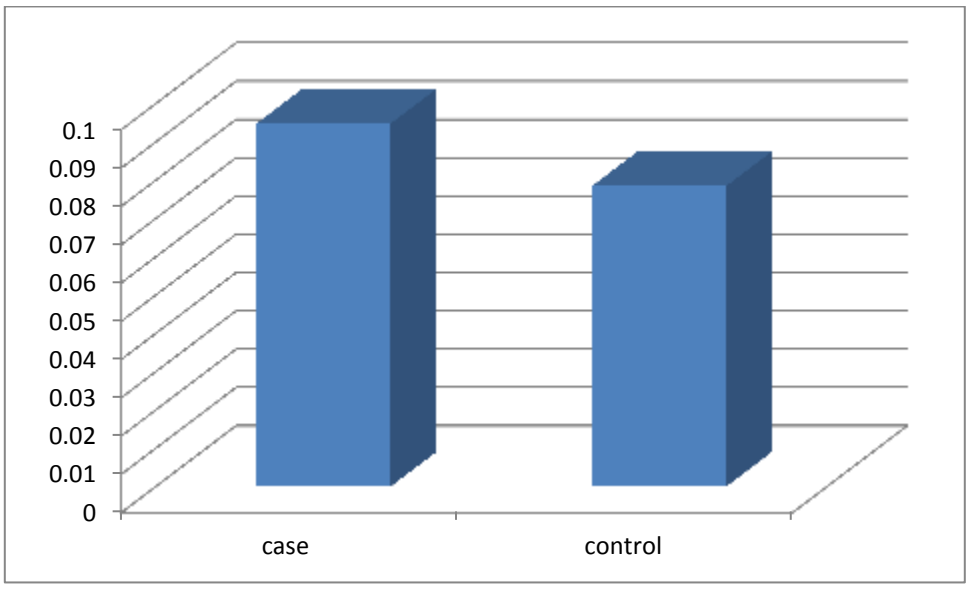


Figure (4_1): the mean and Standard deviation of plasma chromium level in case and control group

- **Table (4-2) The mean of plasma chromium in diabetic patients according to gender:**

Gender	**Frequency**	**percent %**	**Mean (mg\dl)**	**p.value**
Male	18	36%	0.1039	0.196
Female	32	64%	0.0897	

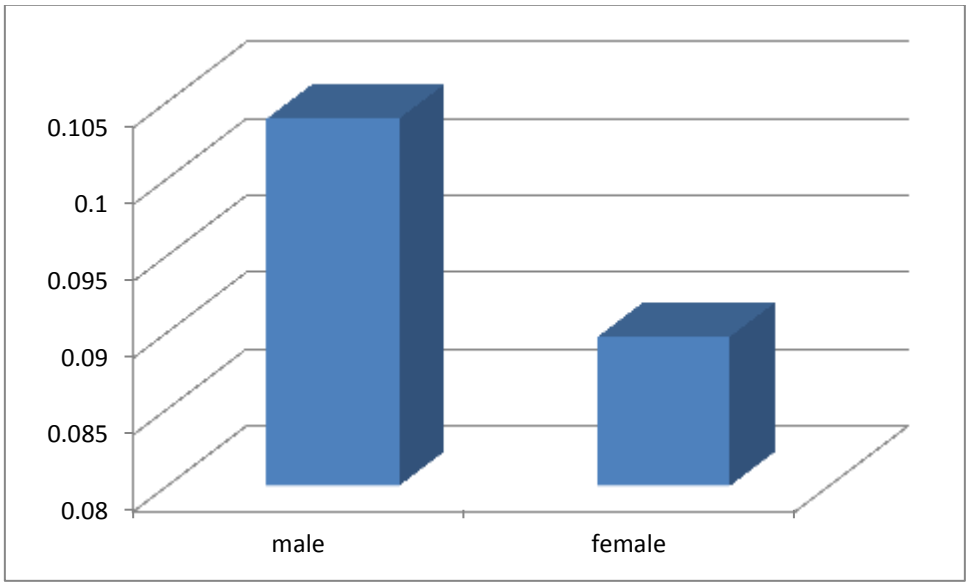


Figure (4_2): The mean of plasma chromium in diabetic patients according to gender

Table (4- 3): The mean of plasma chromium according to the types of diabetes mellitus:

Type of diabetes mellutis	Frequency	percent %	Mean (mg\dl)	p.value
Type 1	5	10%	0.0940	0.960
Type 2	45	90%	0.0949	

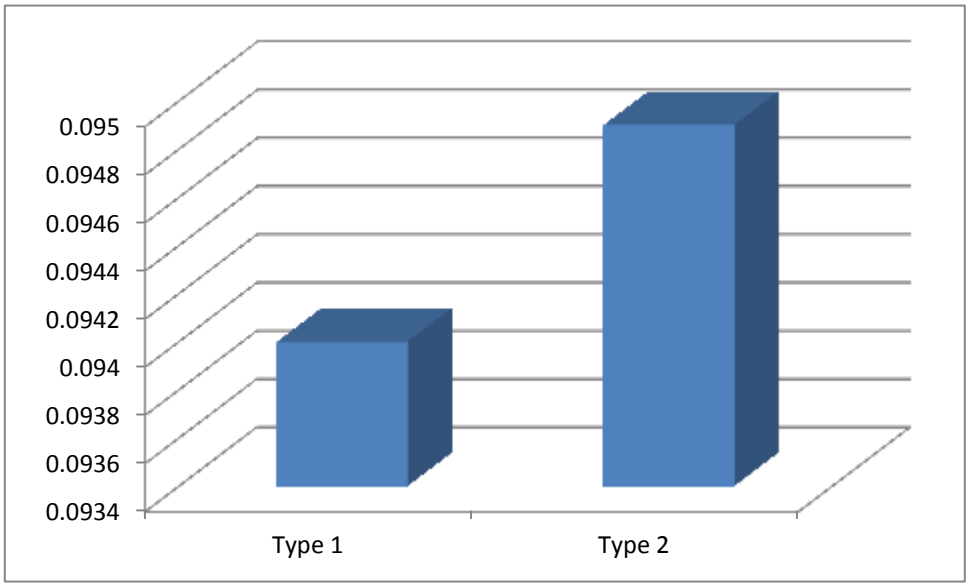


Figure (4_3): The mean of plasma chromium according to the types of diabetes mellitus

Table (4-4) the mean of plasma chromium according to duration of diabetes mellitus:

Duration per years	Frequency	Percentage %	Mean (mg\dl)	p.value
< 3	15	30%	0.090	0.541
4 – 9	15	30%	0.090	
>10	20	40%	0.102	

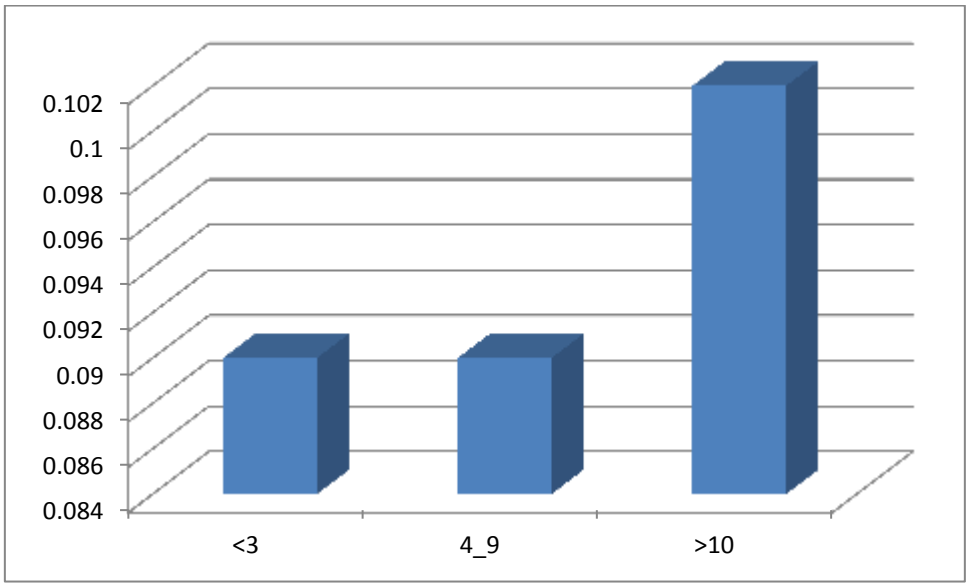


Figure (4_4): the mean of plasma chromium according to duration of diabetes mellitus

Chapter Five

Discussion
Conclusion
Recommendations

5.1. Discussion

The study was conducted in Shendi locality in the period from March to August 2018 to measure the effect of diabetes mellitus on plasma chromium level. 64% of study population was female while 36% was male, 10% were type 1 diabetes mellitus and 90% were type 2 diabetes mellitus.

The mean of plasma chromium in diabetic patient was (0.0948 mg/dl), while it was (0.0785 mg/dl) in control group with P. value of (0.160) which is more than 0.05 that confirm there was a statistically insignificant variation between the mean of plasma chromium in diabetic patients and control group as illustrated in (table4.1),Our finding was disagree with previous reports done by Ekmekcioglu C, et al. 2001 who stated that: there is association between plasma chromium and diabetes mellitus.(**Ekmekcioglu, Prohaska et al. 2001**).

Regarding to the gender the mean of plasma chromium in male with diabetes was (0.1039 mg/dl) while it was (0.0897 mg/dl) in female with diabetes that mean gender is not affect on plasma chromium in patient with diabetes mellitus (table 4.2),and this disagree with previous study done by **(Kazi, Afridi et al. 2008)**.

According to the type of diabetes mellitus the mean of plasma chromium in type1 diabetes mellitus was (0.0940 mg/dl) while it was (0.0949mg/dl) in type 2 diabetes mellitus, the p. value more than 0.05 that mean the type of diabetes mellitus is not affect on plasma chromium in patient with (table 4.3),and this disagree with previous study done by (**Ekmekcioglu, Prohaska et al. 2001**).

In table (4-4) show the effect of duration on chromium level which found the mean of people from 1_3 years is 0.0900 mg/dl and from 4_9 years is 0.0900 mg/dl and in more than 10 is 0.1020 mg/dl and the p value is more than 0.05 that mean insignificant, and this disagree with previous study done by (**Ekmekcioglu, Prohaska et al. 2001**) **.**

5.2 Conclusion

On the basis of the study results we can conclude the following:

- Insignificant Variation in level of plasma chromium between case group and control group that mean the diabetes mellitus not affect in the chromium level.
- Insignificant variations that mean the type of diabetes mellitus not affect the chromium level.
- Insignificant variation between the duration of disease and chromium level.
- Insignificant variation between mean of chromium level in male and female that mean gender it not affect plasma chromium level.

5.3. Recommendations

1- Regular visiting of diabetes health centers and monitoring of serum glucose level

2- Improve health education programs about diabetes and it is complication for population including different age group. Especially to patient with diabetes.

3- Diabetic patients should eat foods containing trace elements, to prevent diabetic complication from occurring.

4- In soon future other study can conducted with large sample size and the study can compose control and in control patients.

Chapter Six

References
Appendix

References:

Al-Fartusie, F. S. and S. N. Mohssan (2017). "Essential Trace Elements and Their Vital Roles in Human Body." Indian Journal of Advances in Chemical Science **5**(3): 127-136.

Arneson, W. L. and J. M. Brickell (2007). Clinical Chemistry: a laboratory perspective, FA Davis.

Association, A. D. (2014). "Diagnosis and classification of diabetes mellitus." Diabetes care **37**(Supplement 1): S81-S90.

Basaki, M., M. Saeb, et al. (2012). "Zinc, copper, iron, and chromium concentrations in young patients with type 2 diabetes mellitus." Biological Trace Element Research **148**(2): 161-164.

Bishop, M. L., E. P. Fody, et al. (2010). Clinical chemistry: techniques, principles, correlations, Wolters Kluwer Health/Lippincott Williams & Wilkins.

Blake, S. (2008). Vitamins and minerals demystified, McGraw-Hill.

Crook, M. A. (2012). Clinical Biochemistry and Metabolic Medicine Eighth Edition, CRC Press.

Ekmekcioglu, C., C. Prohaska, et al. (2001). "Concentrations of seven trace elements in different hematological matrices in patients with type 2 diabetes as compared to healthy controls." Biological Trace Element Research **79**(3): 205-219.

Fowler, M. J. (2008). "Microvascular and macrovascular complications of diabetes." Clinical diabetes **26**(2): 77-82.

Hajra, B., S. A. Orakzai, et al. (2016). "INSULIN SENSITIVITY TO TRACE METALS (CHROMIUM, MANGANESE) IN TYPE 2 DIABETIC PATIENTS AND NON DIABETIC INDIVIDUALS." Journal of Ayub Medical College Abbottabad **28**(3): 534-536.

Kazi, T. G., H. I. Afridi, et al. (2008). "Copper, chromium, manganese, iron, nickel, and zinc levels in biological samples of diabetes mellitus patients." Biological Trace Element Research **122**(1): 1-18.

Lipko, M. and B. Debski (2018). "Mechanism of insulin-like effect of chromium (III) ions on glucose uptake in C2C12 mouse myotubes involves ROS formation." Journal of Trace Elements in Medicine and Biology **45**: 171-175.

Loewen, S. and L. Haas (1991). Complications of diabetes: acute and chronic. Nurse practitioner forum.

Ngala, R. A., M. A. Awe, et al. (2018). "The effects of plasma chromium on lipid profile, glucose metabolism and cardiovascular risk in type 2 diabetes mellitus. A case-control study." PloS one **13**(7): e0197977.

Organization, W. H. (1999). Definition, diagnosis and classification of diabetes mellitus and its complications: report of a WHO consultation. Part 1, Diagnosis and classification of diabetes mellitus, Geneva: World health organization.

Ozougwu, J., K. Obimba, et al. (2013). "The pathogenesis and pathophysiology of type 1 and type 2 diabetes mellitus." Journal of Physiology and Pathophysiology **4**(4): 46-57.

Rother, K. I. (2007). "Diabetes treatment—bridging the divide." The New England journal of medicine **356**(15): 1499.

Shaw, J. C. (1980). "Trace elements in the fetus and young infant: II. Copper, manganese, selenium, and chromium." American Journal of Diseases of Children **134**(1): 74-81.

Wada, O. (2004). "What are Trace Elements?".

Appendixes I

Atomic absorption spectrophotometer

Appendixes II

Questionnaire :

Name: ……

1- Sex: a. Male:( ) b. Female:( )

2- Age: ……

3- Weight: ……

4- Height: ……

5- Body mass index: ……

6- Marital status: a. Single:( ) b. married:( )

7-Education level: a. Educated:( ) b. not educated:( )

8- where do you live: ……

9- type of diabetes: a. Type 1:( ) b.Type 2:( )

10- duration of diabetes: ……

11- hypertensive: a.Yes:( ) b.No:( )

12- heart disease: a.Yes:( ) b.No:( )

13- Exercise: a.Regular:( ) b.Irregular:( ) c.Never:( )

14- Do you smoke: a.Yes:( ) b. No:( )

15-If yes: duration of smoking: ……

16- Are you on a controlled diet: a.Always:( ) b.Sometimes:( ) c.Never:( )

Appendixes III

بسم الله الرحمن الرحيم

إقـرار بالموافقـة

الاسم :----------

العمر :---------- العنوان :----------

أوافق بمحض ارادتى بالمشاركة فى البحث العلمى المتعلق بدراسة الكروم في المرضى السودانين المصابين بالسكري .

الطالبة :فاطمة عوض الكريم محمد الجلال

بعد أن شرح لى بأنه لا يترتب عليه أى أذى جسدى واعلم أن المشاركة في هذا البحث لن تؤثر بأي حال من الأحوال في الرعاية الطبية التي يتلقاها مولودي كما أنه يحق لي بدون ابداء أسباب الإنسحاب من هذا البحث في أي مرحلة من مراحله.

البحث بإشراف :

د.مصعب عمر خالد محمد زين

التوقيع : ---------- التاريخ :----------
