CHAPTER ONE

1. Introduction and Literature Review:

1.1 Introduction:

Hypertension or high blood pressure is a cardiac chronic medical condition in which the systemic arterial blood pressure is elevated. What that means is that the heart has to work harder than it should to pump the blood around the body. Blood pressure involves two measurements, systolic and diastolic. Normal blood pressure is 120/80 mm/Hg. The first figure is the systolic blood pressure, the pressure there is in the arteries when the heart is contracting. The second, or lower figure, is the diastolic blood pressure, which is the pressure in arteries between heart beats. High blood pressure is anything above 140/90 mm/Hg. Hypertension is classified as either primary hypertension or secondary hypertension; about 90–95% of cases are categorized as "primary hypertension," which means high blood pressure with no obvious medical cause. The remaining 5–10% of cases (Secondary hypertension) is caused by other conditions that affect the kidneys, arteries, heart or endocrine system. Persistent hypertension is one of the risk factors for stroke, myocardial infarction, heart failure and arterial aneurysm, and is a leading cause of chronic kidney failure. Moderate elevation of arterial blood pressure leads to shortened life expectancy. Dietary and lifestyle changes can improve blood pressure control and decrease the risk of associated health complications, although drug treatment may prove necessary in patients for whom lifestyle changes prove ineffective or insufficient. In the year 2000 it is estimated that nearly one billion people or 26% of the adult population had hypertension worldwide it has common in both developed 333 million and undeveloped 639 million countries, It more common in blacks and native in American and less in white and Mexican American rates increase with age and is greater in the southeastern united states hypertension is more prevalent in men [though menopause tends to decrease this difference and those of low socioeconomic status. Over 90-95 percent of adult hypertension is essential hypertension one of the most common causes of secondary hypertension is primary aldosteronism. The incidence of exercise hypertension is reported to range from 1-10%. 
1.2 Rational:
Hypertension is one of the most common worldwide diseases afflicting humans and is a major risk factor for stroke, myocardial infarction, vascular disease, and chronic kidney disease. According to the American Heart Association (AHA), approximately 75 million adults in the United States are affected by hypertension, which is defined as a systolic blood pressure (SBP) of 140 mm Hg or more or a diastolic blood pressure (DBP) of 90 mm Hg or more or taking antihypertensive medication.

As previous studies reported and asocial of hypertensive and renal impairment, the aim of this study to assess the plasma calcium and uric acid levels in hypertensive patient and correlation them with the duration of hypertensive and outcomes may be useful to managing hypertensive and it is complications in hypertensive Sudanese patients.
1.3 Literature review:

1.3.1 Blood pressure:

Blood pressure assessment is an integral part of clinical practice. Routinely, a patient’s blood pressure is obtained at every physical examination, including outpatient visits, at least daily when patients are hospitalized, and before most medical procedures. Blood pressure measurements are obtained for a wide variety of reasons, including screening for hypertension, assessing a person’s suitability for a sport or certain occupations, estimating cardiovascular risk and determining risk for various medical procedures. (2)

Blood pressure measurements are also obtained routinely when following a hypertensive patient to assist with tailoring of medications and treatment of hypertension. Finally, blood pressure measurements are an integral part of identifying if a patient is in potential or actual clinical deterioration. (2)

Two methods for measuring a blood pressure exist, the direct and indirect method. The direct method is the criterion standard and consists of using an intra-arterial catheter to obtain a measurement. This method, however, is not practical due to its invasiveness and its inability to be applied to large groups of asymptomatic individuals for hypertension screening. (2)

Therefore, the indirect (noninvasive) method is typically used. The indirect method involves collapsing the artery with an external cuff, providing an inexpensive and easily reproducible way to measure blood pressure. The indirect method can be performed using a manual cuff and sphygmomanometer or with an automated oscillometric device. The manual method requires auscultation of the blood pressure, whereas the automated system depends on oscillometric devices. (2)

1.3.2 Hypertension:

Hypertension is one of the most common worldwide diseases afflicting humans and is a major risk factor for stroke, myocardial infarction, vascular disease, and chronic kidney disease. Despite extensive research over the past several decades, the etiology of most cases of adult hypertension is still unknown, and control of blood pressure is suboptimal in the general population. Due to the associated morbidity and mortality and cost to society, preventing and treating hypertension is an important public health challenge. Fortunately, recent advances and trials in hypertension research are leading to an increased understanding of the pathophysiology
of hypertension and the promise for novel pharmacologic and interventional treatments for this widespread disease.\textsuperscript{(4)}

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Substantial improvements have been made with regard to enhancing awareness and treatment of hypertension. However, a National Health Examination Survey (NHANES) spanning 2005-2006 showed that 29\% of US adults 18 years of age and older were hypertensive; 7\% of hypertensive adults had never been told that they had hypertension.\textsuperscript{(6)}

Furthermore, of those with high blood pressure (BP), 78\% were aware they were hypertensive, 68\% were being treated with antihypertensive agents, and only 64\% of treated individuals had controlled hypertension. In addition, data from NHANES 1999-2006 estimated that 30\% of adults 20 years of age and older have prehypertension, defined as an untreated SBP of 120-139 mm Hg or untreated DBP of 80-89 mmHg.\textsuperscript{(7)}

Data from the Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure (JNC 7), which was released in 2003, were relatively similar to the NHANES data. The JNC 7 noted that approximately 30\% of adults were unaware of their hypertension; up to 40\% of people with hypertension were not receiving treatment; and, of those treated, up to 67\% did not have their BP controlled to less than 140/90.

Hypertension is the most important modifiable risk factor for coronary heart disease (the leading cause of death in North America), stroke (the third leading cause), congestive heart failure, end-stage renal disease, and peripheral vascular disease. Therefore, health care professionals must not only identify and treat patients with hypertension but also promote a healthy lifestyle and preventive strategies to decrease the prevalence of hypertension in the general population.\textsuperscript{(7)}

Most individuals diagnosed with hypertension will have increasing blood pressure (BP) as they age. Untreated hypertension is notorious for increasing the risk of mortality and is often described as a silent killer. Mild to moderate hypertension, if left untreated, may be associated with a risk of atherosclerotic disease in 30\% of people and organ damage in 50\% of people within 8-10 years after onset.\textsuperscript{(7)}
Death from ischemic heart disease or stroke increases progressively as BP increases. For every 20 mm Hg systolic or 10 mm Hg diastolic increase in BP above 115/75 mm Hg, the mortality rate for both ischemic heart disease and stroke doubles.\(^8\)

In the Framingham Heart Study, the age-adjusted risk of congestive heart failure was 2.3 times higher in men and 3 times higher in women when the highest BP was compared to the lowest.\(^9\)

Multiple Risk Factor Intervention Trial (MRFIT) data showed that the relative risk for coronary artery disease mortality was 2.3 to 6.9 times higher for persons with mild to severe hypertension than it was for persons with normal BP.\(^{10}\)

The relative risk for stroke ranged from 3.6 to 19.2. The population-attributable risk percentage for coronary artery disease varied from 2.3 to 25.6%, whereas the population-attributable risk for stroke ranged from 6.8-40 %.\(^{10}\)

The Framingham Heart Study found a 72% increase in the risk of all-cause death and a 57% increase in the risk of any cardiovascular event in patients with hypertension who were also diagnosed with diabetes mellitus.\(^{11}\)

Nephrosclerosis is one of the possible complications of long-standing hypertension. The risk of hypertension-induced end-stage renal disease is higher in black patients, even when blood pressure is under good control. Furthermore, patients with diabetic nephropathy who are hypertensive are also at high risk for developing end-stage renal disease.\(^{11}\)

### 1.3.2.1 Signs and symptoms:

The 2013 joint European Society of Hypertension (ESH) and the European Society of Cardiology (ESC) guidelines recommend that ambulatory blood-pressure monitoring (ABPM) be incorporated into the assessment of cardiovascular risk factors and hypertension.\(^{12}\)

Following the documentation of hypertension, which is confirmed after an elevated blood pressure (BP) on at least 3 separate occasions (based on the average of 2 or more readings taken at each of ≥2 follow-up visits after initial screening), a detailed history should extract the following information: extent of end-organ damage (eg, heart, brain, kidneys, eyes), Assessment of patients’ cardiovascular risk status, Exclusion of secondary causes of hypertension

Patients may have undiagnosed hypertension for years without having had their BP checked. Therefore, a careful history of end-organ damage should be obtained.
1.3.2.2 Classification of hypertension:

Primary (essential) hypertension is the most common form of hypertension, accounting for 90–95% of all cases of hypertension. In almost all contemporary societies, blood pressure rises with aging and the risk of becoming hypertensive in later life is considerable. Hypertension results from a complex interaction of genes and environmental factors. Numerous common genetic variants with small effects on blood pressure have been identified as well as some rare genetic variants with large effects on blood pressure but the genetic basis of hypertension is still poorly understood. Several environmental factors influence blood pressure. Lifestyle factors that lower blood pressure include reduced dietary salt intake, increased consumption of fruits and low fat products, exercise, weight loss and reduced alcohol intake. Stress appears to play a minor role with specific relaxation techniques not supported by the evidence. The possible role of other factors such as caffeine consumption and vitamin D deficiency are less clear cut. Insulin resistance, which is common in obesity and is a component of syndrome X (or the metabolic syndrome), is also thought to contribute to hypertension. Recent studies have also implicated events in early life (for example low birth weight, maternal smoking and lack of breast feeding) as risk factors for adult essential hypertension, although the mechanisms linking these exposures to adult hypertension remain obscure. (12)

Secondary hypertension results from an identifiable cause. Renal disease is the most common secondary cause of hypertension. Endocrines such as Cushing’s syndrome, hyperthyroidism, hypothyroidism, acromegaly, hyperaldosteronism, hyperparathyroidism and pheochromocytoma.

Other causes include obesity, sleep apnea, pregnancy, coarctation of the aorta, excessive liquorice consumption and certain prescription medicines, herbal remedies and illegal drugs. (12)

1.3.2.3 Pathophysiology:

In most people with established essential (primary) hypertension, increased resistance to blood flow (total peripheral resistance) accounting for the high pressure while cardiac output remains normal. There is evidence that some younger people with prehypertension or 'borderline hypertension' have high cardiac output, an elevated heart rate and normal peripheral resistance, termed hyperkinetic borderline hypertension. These individuals develop the typical features of established essential hypertension in later life as their cardiac output falls and peripheral resistance rises with age. Whether this pattern is typical of all people who ultimately develop
hypertension is disputed. The increased peripheral resistance in established hypertension is mainly attributable to structural narrowing of small arteries and arterioles, although a reduction in the number or density of capillaries may also contribute. Hypertension is also associated with decreased peripheral venous compliance which may increase venous return, increase cardiac preload and, ultimately, cause diastolic dysfunction. Whether increased active vasoconstriction plays a role in established essential hypertension is unclear.\\(^{13}\)

Pulse pressure (the difference between systolic and diastolic blood pressure) is frequently increased in older people with hypertension. This can mean that systolic pressure is abnormally high, but diastolic pressure may be normal or low a condition termed isolated systolic hypertension. The high pulse pressure in elderly people with hypertension or isolated systolic hypertension is explained by increased arterial stiffness, which typically accompanies aging and may be exacerbated by high blood pressure.\\(^{14}\)

Many mechanisms have been proposed to account for the rise in peripheral resistance in hypertension. Most evidence implicates either disturbances in renal salt and water handling (particularly abnormalities in the intrarenal renin-angiotensin system) and/or abnormalities of the system. These mechanisms are not mutually exclusive and it is likely that both contribute to some extent in most cases of essential hypertension. It has also been suggested that endothelial dysfunction and vascular inflammation may also contribute to increased peripheral resistance and vascular damage in hypertension.\\(^{15}\)

1.3.2.4 Management:

1.3.2.4.1 Lifestyle modifications:

The first line of treatment for hypertension is identical to the recommended preventive lifestyle changes and includes dietary changes, physical exercise, and weight loss. These have all been shown to significantly reduce blood pressure in people with hypertension. Their potential effectiveness is similar to using a single medication. If hypertension is high enough to justify immediate use of medications, lifestyle changes are still recommended in conjunction with medication.\\(^{16}\)

Dietary change such as a low sodium diet is beneficial. A long term (more than 4 weeks) low sodium diet in Caucasians is effective in reducing blood pressure, both in people with hypertension and in people with normal blood pressure. Also, the DASH diet, a diet rich in nuts,
whole grains, fish, poultry, fruits and vegetables lowers blood pressure. A major feature of the plan is limiting intake of sodium although the diet is also rich in potassium, magnesium, calcium, as well as protein. Different programs aimed to reduce psychological stress such as biofeedback, relaxation or transcendental meditation may be reasonable add-ons to other treatment to reduce hypertension. However, overall efficacy is not greater than health education, with evidence being generally of low quality.\textsuperscript{(16)}

1.3.2.4.2 Medications:
Several classes of medications, collectively referred to as antihypertensive drugs, are currently available for treating hypertension. Use should take into account the person's cardiovascular risk (including risk of myocardial infarction and stroke) as well as blood pressure readings, in order to gain a more accurate picture of the person's cardiovascular profile. Evidence in those with mild hypertension (SBP less than 160 mmHg and/or DBP less than 100 mmHg) and no other health problems does not support a reduction in the risk of death or rate of health complications from medication treatment. Medications are not recommended for people with prehypertension or high normal blood pressure.\textsuperscript{(17)}

1.3.2.5 Complications of Hypertension:
Complications of hypertension are clinical outcomes that result from persistent elevation of blood pressure. Hypertension is a risk factor for all clinical manifestations of atherosclerosis since it is a risk factor for atherosclerosis itself. It is an independent predisposing factor for heart failure, coronary artery disease, stroke, renal disease and peripheral arterial disease. It is the most important risk factor for cardiovascular morbidity and mortality in industrialized countries.\textsuperscript{(20)}

1.3.2.5.1 Complications affecting the heart:
Hypertensive heart disease is the result of structural and functional adaptations leading to left ventricular hypertrophy, diastolic dysfunction, CHF, abnormalities of blood flow due to atherosclerotic coronary artery disease and microvascular disease, and cardiac arrhythmias. Individuals with left ventricular hypertrophy are at increased risk for stroke, CHF, and sudden death. Aggressive control of hypertension can regress or reverse left ventricular hypertrophy and reduce the risk of cardiovascular disease. Left ventricular hypertrophy are seen in 25% of the hypertensive patients and can easily be diagnosed by using echocardiography. Underlying
mechanisms of hypertensive left ventricular hypertrophy are of 2 types: mechanical, mainly leading to myocyte hypertrophy; neuro-hormonal, mainly resulting in a fibroblastic proliferation. Abnormalities of diastolic function, ranging from asymptomatic heart disease to overt heart failure are common in hypertensive patients. Patients with diastolic heart failure have a preserved ejection fraction, which is a measure of systolic function. Diastolic dysfunction is an early consequence of hypertension-related heart disease and is exacerbated by left ventricular hypertrophy and ischemia.\(^{21}\)

1.3.2.5.2 Complications affecting the brain:
Hypertension is an important risk factor for brain infarction and hemorrhage. Approximately 85% of strokes are due to infarction and the remainder is due to hemorrhage, either intracerebral hemorrhage or subarachnoid hemorrhage. The incidence of stroke rises progressively with increasing blood pressure levels, particularly systolic blood pressure in individuals >65 years. Treatment of hypertension convincingly decreases the incidence of both ischemic and hemorrhagic strokes. \(^{22}\)

1.3.2.5.3 Complications affecting the eye:
Hypertensive retinopathy is a condition characterized by a spectrum of retinal vascular signs in people with elevated blood pressure. The retinal circulation undergoes a series of pathophysiological changes in response to elevated blood pressure. In the initial, vasoconstrictive stage, there is vasospasm and an increase in retinal arteriolar tone owing to local autoregulatory mechanisms. This stage is seen clinically as a generalized narrowing of the retinal arterioles. Persistently elevated blood pressure leads to intimal thickening, hyperplasia of the media wall, and hyaline degeneration in the subsequent, sclerotic, stage. This stage corresponds to more severe generalized and focal areas of arteriolar narrowing, widening and accentuation of the central light reflex. \(^{23}\)

1.3.2.5.4 Complications affecting the kidneys:
Hypertension is a risk factor for renal injury and ESRD. Renal risk appears to be more closely related to systolic than to diastolic blood pressure, and black men are at greater risk than white men for developing ESRD at every level of blood pressure, the atherosclerotic, hypertension-related vascular lesions in the kidney primarily affect the preglomerular arterioles, resulting in ischemic changes in the glomeruli and post glomerular structures. Glomerular injury may also be a consequence of direct damage to the glomerular capillaries due to glomerular hyperperfusion.
Glomerular pathology progresses to glomerulosclerosis, and eventually the renal tubules may also become ischemic and gradually atrophic. The renal lesion associated with malignant hypertension consists of fibrinoid necrosis of the afferent arterioles, sometimes extending into the glomerulus, and may result in necrosis. Clinically, macroalbuminuria (a random urine albumin/creatinine ratio > 300 mg/g) or macroalbuminuria (a random urine albumin/creatinine ratio 30–300 mg/g) are early markers of renal injury. These are also risk factors for renal disease progression and for cardiovascular disease. (24)
1.3.3 Uric acid:

Uric acid (urate) is an organic compound of carbon, nitrogen, oxygen, and hydrogen with the formula $\text{C}_5\text{H}_4\text{N}_4\text{O}_3$, the enzyme xanthine oxidase makes uric acid from xanthine and hypoxanthine, which in turn are produced from other purines. Xanthine oxidase is a large enzyme whose active site consists of the metal molybdenum bound to sulfur and oxygen. Within cells, xanthine oxidase can exist as xanthine dehydrogenase and xanthine oxireductase, which has also been purified from bovine milk and spleen extracts. Uric acid is released in hypoxic conditions.\(^{(25)}\)

In humans and higher primates, uric acid is the final oxidation (breakdown) product of purine metabolism and is excreted in urine. In most other mammals, the enzyme uricase further oxidizes uric acid to allantoin. The loss of uricase in higher primates parallels the similar loss of the ability to synthesize ascorbic acid, leading to the suggestion that urate may partially substitute for ascorbate in such species. Both uric acid and ascorbic acid are strong reducing agents (electron donors) and potent antioxidants. In humans, over half the antioxidant capacity of blood plasma comes from uric acid.\(^{(26)}\)

The Dalmatian dog has a genetic defect in uric acid uptake by the liver and kidneys, resulting in decreased conversion to allantoin, so this breed excretes uric acid, and not allantoin, in the urine.

In humans, about 70% of daily uric acid disposal occurs via the kidneys, and in 5-25% of humans, impaired renal (kidney) excretion leads to hyperuricemia.\(^{(26)}\)

1.3.3.1 Source of uric acid:

In many instances, people have elevated uric acid levels for hereditary reasons. Diet may also be a factor. In humans, purines are excreted as uric acid. Purines are found in high amounts in animal food products, such as liver and sardines. A moderate amount of purine is also contained in beef, pork, poultry, fish and seafood, asparagus, cauliflower, spinach, mushrooms, green peas, lentils, dried peas, beans, oatmeal, wheat bran, and wheat germ.\(^{(26)}\)

Examples of high purine sources include: sweetbreads, anchovies, sardines, liver, beef kidneys, brains, meat extracts (e.g., Oxo, Bovril), herring, mackerel, game meats, beer, and gravy.

Moderate intake of purine-containing vegetables is not associated with an increased risk of gout.

Uric acid, produced mostly in the liver is the end product of dietary and endogenous purine metabolism Ribose-5-phosphate, derived from glycidic metabolism, is converted to phosphoribosyl pyrophosphate (PRPP) via the PRPP synthetase and then to
inosinemonophosphate. This intermediate compound yields adenosine monophosphate (AMP) and guanosine monophosphate, the purinic nucleotides used in DNA and RNA synthesis, as well as inosine. The latter is converted by the purine nucleoside phosphorylase to hypoxanthine. Xanthine oxidase (XO), an enzyme inhibited by allopurinol, converts hypoxanthine to xanthine and subsequently xanthine to uric acid. Hypoxanthine and guanine may enter a salvage pathway through the activity of hypoxanthine–guanine phosphoribosyltransferase (HGPRT), an enzyme that reconverts these purine bases into their respective nucleotides. Animals that possess an active urate oxidase (uricase) can transform uric acid into a more soluble molecule, allantoin. In humans, however, uricase is inactive as a result of a nonsense mutation. Key enzymes that cause abnormal uric acid levels include: PRPP synthetase, purine nucleoside phosphorylase, XO and HGPRT. (27)

1.3.3.2 Excretion:

In humans, the excretory processes are the formation of urine in the kidneys and formation of carbon dioxide molecules as result of respiration, which is then exhaled from the lung. These waste products are eliminated by urination and exhalation respectively. In urination, hormone control over excretion occurs in the distal tubules of kidneys as directed by the hypothalamus. (28) Although uric acid provides an efficient means of eliminating nitrogen, it is poorly soluble in biological fluids. With a functional pKa of 5.75 in blood, uric acid exists predominantly as urate anion at physiologic pH; however, it exists mostly in the uric acid form in urine at a pH of approximately 5–6, which affects its solubility and transport. The saturation level of monosodium urate in human plasma is approximately 7 mg/dl; hence, the definition of hyperuricemia is usually a plasma uric acid of >7 mg/dl.

Uric acid must be excreted continuously to prevent toxic accumulation. Human tissues have a very limited ability to metabolize urate, which must be eliminated by the gut and kidney to maintain homeostasis. Approximately 75 % of daily urate is excreted by the kidney. Colonic bacteria degrade uric acid to allantoin. (28)

Renal excretion of uric acid consists of four steps: (1) glomerular filtration, (2) presecretory reabsorption, (3) secretion and (4) postsecretory reabsorption. Nearly all of the urate is filtered through the glomerulus, with most of the filtered urate (99 %) being reabsorbed in the early S1 segment of the proximal tubule (presecretory reabsorption). This is followed by tubular secretion in the S2 segment of the proximal tubule that returns approximately 50 % of the filtered
urate into the tubular lumen. However, the majority (40%) of the secreted urate undergoes postsecretory reabsorption that occurs predominately in the last segment (S3) of the proximal tubule. (29)

The molecular mechanisms that control urate transport are not fully understood. Genome-wide association studies have revealed the complex interplay of membrane transporters involved in urate metabolism. Excretion is dependent on a number of urate transporters, including urate anion transporter 1 (URAT1), organic anion transporters (OAT1 and OAT3) and ATP-(adenosine triphosphate) dependent urate export transporters multidrug resistance protein 4. Transporter URAT1, encoded by the SLC22A12 gene and localized on the luminal membrane of the proximal tubule, is responsible for the reabsorption of urate after glomerular filtration. Intracellular accumulation of organic anions, such as lactate, acetoacetate, hydroxybutyrate and succinate, favors the reabsorption of urate in exchange for these anions which translocate to the tubular lumen. This process provides the basis of hyperuricemia associated with increased levels of these anions. Glucose transporter 9 (Glut9), encoded by the SLC2A9 gene, is a fructose transporter that mediates urate reabsorption from the tubular cell to the circulation. The reabsorption of urate by Glut9 and URAT is inhibited by probencid and benz bromarone. URAT1 is also inhibited by losartan, an angiotensin II receptor blocker. (29)

1.3.3.3 Uric acid Levels:

Concentrations of uric acid might have a great impact on the monitoring, diagnosis, prognosis and therapy of several renal disorders. Serum urate concentrations in most children range from 3 to 4 mg/dl. During male puberty, levels begin to rise. Throughout adolescence, fractional excretion of urate by the kidney declines to that found in adults, and there is a substantial increase in body mass. Both changes contribute to an increase in uric acid levels. During early adolescence, when girls weigh more than age-matched boys, girls have slightly higher uric acid levels than boys. By mid adolescence, both weight and uric acid levels in boys exceed those in girls. This gender discrepancy in serum uric acid levels persists until menopause and may be due to the hypouricemic effect of estrogens. (30)

Normal values range between 3.5 and 7.2 mg/dL. (30)

1.3.2.3.1 Hyperuricemia:

Clinically significant hyperuricemia may result from either overproduction, decreased renal uric acid excretion or a combination of both mechanisms. Increased production Any disease process
with high cell turnover, with increased purine metabolism and uric acid production may produce hyperuricemia. Accelerated purine degradation can result from rapid cell proliferation and turnover (such as leukemias and lymphomas) or from cell death (rhabdomyolysis, cytotoxic therapy). Glycogenosis types III, IV and VII can result in hyperuricemia from excessive degradation of skeletal muscle ATP. In addition, a small percentage of overproducers have enzymatic defects that account for hyperuricemia. These include a complete deficiency of HGPRT as in Lesch–Nyhan syndrome, partial deficiency of HGPRT (Kelley–Seegmiller syndrome) and increased PRPP activity. Furthermore the role played by diet in hyperuricemia has not yet been fully clarified; nonetheless, high intake of fructose-rich industrialized food and high alcohol intake (particularly beer) seem to influence uricemia. High fructose intake can lead to increased uric acid production; it causes fructose to be rapidly phosphorylated by fructokinase to fructose 1-phosphate leading to low concentrations of intracellular phosphate. This limits ATP formation and results in the generation of adenosine diphosphate or AMP, which are further metabolized to uric acid. Beer intake also leads to hyperuricemia due to its alcoholic content and high quality purine.\(^{(31)}\)

Decreased renal clearance Altered uric acid excretion can result from decreased glomerular filtration, decreased tubular secretion or enhanced tubular reabsorption. While decreased urate filtration may not cause primary hyperuricemia, it can contribute to the hyperuricemia of renal insufficiency. Decreased tubular secretion of urate occurs in patients with acidosis (e.g. diabetic ketoacidosis, ethanol or salicylate intoxication, starvation ketosis). The organic acids that accumulate in these conditions compete with urate for tubular secretion. Finally, enhanced reabsorption of uric acid is responsible for the hyperuricemia observed with diuretic therapy and diabetes insipidus.\(^{(32)}\)

FJHN and autosomal-dominant medullary cystic kidney disease (ADMCKD) are more common but less well-defined hyperuricemic conditions resulting from a decrease in the FEUA with normal urate production. FJHN is an autosomal dominant disorder characterized by early onset hyperuricemia, decreased FEUA and progressive interstitial nephropathy. ADMCKD is characterized by the presence of medullary cysts. About one-third of patients with FJHN and ADMCKD have mutations in the gene for uromodulin, which encodes uromodulin/Tamm–Horsfall.\(^{(32)}\)
Gout

Excess serum accumulation of uric acid in the blood can lead to a type of arthritis known as gout. This painful condition is the result of needle-like crystals of uric acid precipitating in joints, capillaries, skin, and other tissues. Kidney stones can also form through the process of formation and deposition of sodium urate microcrystals. (33)

A study found that men that drink two or more sugar-sweetened beverages a day have an 85% higher chance of developing gout than those that drank such beverages infrequently. (34)

Gout can occur where serum uric acid levels are as low as 6 mg/dL (~357 µmol/L), but an individual can have serum values as high as 9.6 mg/dL (~565 µmol/L) and not have gout.

One treatment for gout, in the 19th century, had been administration of lithium salts lithium urate is more soluble. Today, inflammation during attacks is more commonly treated with NSAIDs or corticosteroids, and urate levels are managed with allopurinol. Allopurinol, developed over 30 years ago by Elion et al., weakly inhibits xanthine oxidase. It is an analog of hypoxanthine that is hydroxylated by xanthine oxireductase at the 2-position to give oxipurinol. Oxipurinol has been supposed to bind tightly to the reduced molybdenum ion in the enzyme and, thus, inhibits uric acid synthesis. (35)

Lesch-Nyhan syndrome, an extremely rare inherited disorder, is also associated with very high serum uric acid levels. Spasticity, involuntary movement, and cognitive retardation as well as manifestations of gout are seen in cases of this syndrome. (36)

Although uric acid can act as an antioxidant, excess serum accumulation is often associated with cardiovascular disease. It is not known whether this is causative (e.g., by acting as a prooxidant) or a protective reaction taking advantage of urate's antioxidant properties. The same may account for the putative role of uric acid in the etiology of stroke.

The association of high serum uric acid with insulin resistance has been known since the early part of the 20th century, nevertheless, recognition of high serum uric acid as a risk factor for diabetes has been a matter of debate. In fact, hyperuricemia has always been presumed to be a consequence of insulin resistance rather than its precursor. However, a prospective follow-up study showed high serum uric acid is associated with higher risk of type 2 diabetes, independent of obesity, dyslipidemia, and hypertension. (37)

Hyperuricemia is associated with components of metabolic syndrome. A study has suggested fructose-induced hyperuricemia may play a pathogenic role in the metabolic syndrome. This is
consistent with the increased consumption in recent decades of fructose-containing beverages (such as fruit juices and soft drinks sweetened with sugar and high-fructose corn syrup) and the epidemic of diabetes and obesity.\(^{(38)}\)

Saturation levels of uric acid in blood may result in one form of kidney stones when the urate crystallizes in the kidney. These uric acid stones are radiolucent and so do not appear on an abdominal plain X-ray, and thus their presence must be diagnosed by ultrasound for this reason. Very large stones may be detected on X-ray by their displacement of the surrounding kidney tissues.\(^{(39)}\)

Uric acid stones, which form in the absence of secondary causes such as chronic diarrhea, vigorous exercise, dehydration, and animal protein loading, are felt to be secondary to obesity and insulin resistance seen in metabolic syndrome. Increased dietary acid leads to increased endogenous acid production in the liver and muscles, which in turn leads to an increased acid load to the kidneys. This load is handled more poorly because of renal fat infiltration and insulin resistance, which are felt to impair ammonia excretion (a buffer). The urine is, therefore, quite acidic, and uric acid becomes insoluble, crystallizes and stones form. In addition, naturally present promoter and inhibitor factors may be affected. This explains the high prevalence of uric stones and unusually acidic urine seen in patients with type 2 diabetes. Uric acid crystals can also promote the formation of calcium oxalate stones.\(^{(39)}\)

1.3.3.3.2 Hypouricemia:

Hypouricemia is defined by a serum uric acid level of <2.0mg/dl. It is a marker for primary or secondary tubulopathy and other underlying illness and can be induced by decreased production or increased urinary excretion.\(^{(40)}\)

Lower serum values of uric acid have been associated with multiple sclerosis (MS). MS patients have been found to have serum levels ~194 μmol/L, with patients in relapse averaging ~160 μmol/L and patients in remission averaging ~230 μmol/L.

Serum uric acid in healthy controls was ~290 μmol/L. Conversion factor 1 mg/dL=59.48 μmol/L.\(^{(43)}\)

A 1998 study completed a statistical analysis of 20 million patient records, comparing serum uric acid values in patients with gout and patients with multiple sclerosis. Almost no overlap between the groups was found.
Uric acid has been successfully used in the treatment and prevention of the animal (murine) model of MS. A 2006 study found elevation of serum uric acid values in multiple sclerosis patients, by oral supplementation with inosine, resulted in lower relapse rates, and no adverse effects.\(^{(44)}\) Correcting low or deficient zinc levels can help elevate serum uric acid. Inosine can be used to elevate uric acid levels. Zn inhibits Cu absorption, helping to reduce the high Cu/Fe in some people with hypouricemia. Fe supplements can ensure adequate Fe reserves (ferritin above 25 ng/dl), also correcting the high Cu/Fe.\(^{(45)}\) Uric acid may be a marker of oxidative stress, and may have a potential therapeutic role as an antioxidant. On the other hand, like other strong reducing substances such as ascorbate, uric acid can also act as a prooxidant. Thus, it is unclear whether elevated levels of uric acid in diseases associated with oxidative stress such as stroke and atherosclerosis are a protective response or a primary cause.\(^{(46)}\)

1.3.3.4 Uric acid and hypertension:

Uric acid is implicated in the development of hypertension and cardiovascular disease.\(^{(47)}\) Animal models have demonstrated that hyperuricemia induces an increase in blood pressure, probably due to a chain of events including activation of the renin–angiotensin system and suppression of nitric oxide, leading to increased systemic vascular resistance, followed by uric acid-mediated vasculopathy of the renal afferent arterioles, resulting in a late sodium-sensitive hypertension. Interestingly, high levels of serum uric acid are associated with elevated blood pressure in healthy U.S. adolescents. Small clinical trials, performed in adolescents with newly diagnosed essential hypertension, demonstrate that reduction of serum uric acid can reduce blood pressure. Furthermore, uric acid is associated with metabolic syndrome and is a risk factor for cardiovascular disease.\(^{(48)}\) It is speculated that uric acid is one of the determinants of metabolic syndrome. Moreover, fructose can lead to increased uric acid production, and animal studies have revealed that fructose intake can result in elevated blood pressure as well as features of metabolic syndrome. Additional prospective studies and clinical trials are needed to determine if uric acid is just a marker in a multifactorial metabolic pathway or a causal factor of hypertension, and thus a potential screening and treatment target.\(^{(49)}\)
1.3.4 Calcium:
Calcium is the fifth most abundant element and is the most prevalent cation in the human body. Approximately 1-1.3 kg of calcium can be found in a healthy adult, 99% of which is in the form of hydroxyapatite in the skeleton, the remaining 1% is contained in the extracellular fluid (ECF). Serum (plasma) calcium exists in 3 distinct forms. Approximately 15% is complexed calcium bound to organic and inorganic anions, 40% is bound to albumin, and the remaining 45% circulates as free ionized calcium. (50)

To keep bones strong, your body is constantly breaking down old bone cells and growing new ones, the same way it sheds and replaces skin cells. To fuel bone growth, keep bone density strong, and prevent osteoporosis, you need a good supply of calcium from dairy products and other foods. But you also need enough vitamin D. Without it, you could drink milk all day and the calcium in it wouldn't do you much good. Vitamin D is key in absorbing calcium from the food you eat calcium that would otherwise get sent out of the body as waste.

Your body doesn't make calcium on its own. The best way to get more calcium is to improve your diet. You already know that dairy products such as milk, cheese, and yogurt are good sources of calcium for those who don't have lactose or other dairy intolerance. (50)

Calcium is the most abundant mineral in the human body. The average adult body contains in total approximately 1 kg, 99% in the skeleton in the form of calcium phosphate salts. The extracellular fluid (ECF) contains approximately 22.5 mmol, of which about 9 mmol is in the plasma. Approximately 500 mmol of calcium is exchanged between bone and the ECF over a period of twenty-four hours. (50)

Calcium has several main functions in the body. Calcium acts structurally as supporting material in bones as calcium phosphate. Calcium is also involved in cellular signalling pathways. Intracellular calcium functions as a second messenger in the secretion of many hormones and neurotransmitters. For instance, the influx of calcium into the neuron causes the release of Acetylcholine from pre-synaptic terminals into the neural synapse. Calcium also acts as an intracellular permeation regulator and mediator of muscle contraction. Calcium acts in the contraction of muscles by removing the Triosephosphateisomerase (TPI) subunit from Myosin heads, which has ATPase activity. Calcium also acts as an enzyme cofactor for some clotting factors (enzymes) in the coagulation cascade. (50)
1.3.4.1 Absorption, excretion and regulation:

About 25 mmol of calcium enters the body in a normal diet. Of this, about 40% (10 mmol) is absorbed in small intestine, and 5 mmol leaves the body in feces, netting 5 mmol of calcium a day. (51) Calcium is absorbed across the intestinal brush border membrane, passing through ion channels such as TRPV6. Calbindin is a vitamin D-dependent calcium-binding protein inside intestinal epithelial cells which functions together with TRPV6 and calcium pumps (PMCA1) in the basal membrane to actively transport calcium into the body. Active transport of calcium occurs primarily in the duodenum portion of the intestine when calcium intake is low; and through passive paracellular transport occurs in the jejunum and ileum parts when calcium intake is high, independent of Vitamin D level. (52)

The kidney excretes 250 mmol a day in pro-urine, and resorbs 245 mmol, leading to a net loss in the urine of 5 mmol/d. In addition to this, the kidney processes Vitamin D into calcitriol, the active form that is most effective in assisting intestinal absorption. Both processes are stimulated by parathyroid hormone.

Although calcium flow to and from the bone is neutral, about 5 mmol is turned over a day. Bone serves as an important storage point for calcium, as it contains 99% of the total body calcium. Calcium release from bone is regulated by parathyroid hormone. Calcitonin stimulates incorporation of calcium in bone, although this process is largely independent of calcitonin.

Low calcium intake may also be a risk factor in the development of osteoporosis. In one meta-analysis, the authors found that fifty out of the fifty-two studies that they reviewed showed that calcium intake promoted better bone balance. With a better bone balance, the risk of osteoporosis is lowered. (53)

Calcium levels are tightly regulated in the body. Calcium regulation is primarily controlled by parathyroid hormone (PTH), vitamin D, and calcitonin.

1. Parathyroid hormone is a hormone produced by the parathyroid glands, which are four small glands that surround the thyroid and are found in the anterior part of the lower neck.
2. Vitamin D is obtained through a process that begins with sun exposure to the skin, the process then continues in the liver and kidneys. Vitamin D can also be found in foods such as eggs and dairy products.
3. Calcitonin is produced in specialized cells in the thyroid gland.
Together, these three hormones act on the bones, the kidneys, and the GI tract to regulate calcium levels in the bloodstream.

Primarily calcium is regulated by the actions of 1,25-Dihydroxycholecalciferol (the biologically active form of Vitamin D), parathyroid hormone (PTH), calcitonin and direct exchange with the bone matrix. Plasma calcium levels are regulated by hormonal and non-hormonal mechanisms. The short term control that prevents calcium spiking in the serum is absorption by the bone matrix after the ingestion of substantial amounts of calcium. After about an hour, PTH will be released and not peak for about 8 hours. The PTH is, over time, a very potent regulator of plasma calcium, and controls the conversion of vitamin D into its active form in the kidney. The parathyroid glands are located behind the thyroid, and produce parathyroid hormone in response to low blood calcium levels. (51)

The parafollicular cells of the thyroid produce calcitonin in response to high calcium levels, which decreases blood calcium levels, but its significance is much smaller than that of PTH (54)

1.3.4.2 Clinical Applications:
Assessing the total calcium level is part of a routine health screening, included in the comprehensive metabolic panel and basic metabolic panel, and is used to measure both ionized calcium and bound calcium. (51)

Measurement of the total calcium alone may sometimes be misleading, since this measurement can change without alteration in the ionized calcium concentration. In hypoalbuminemia, although ionized calcium levels remain normal, total calcium levels decrease. The equation used to measure corrected calcium in cases of hyperalbuminemia/hypoalbuminemia is as follows:

\[ \text{Corrected (Ca)} = \text{Measured total (Ca)} + (0.8 \times [4.5 - \text{alb}]) \]

Additionally, in patients who have chronic kidney disease and low serum bicarbonate levels, a low serum albumin level, or both, it is preferable to measure the ionized free calcium rather than the total calcium in order to diagnose hypocalcemia or hypercalcemia. (51)

1.3.2.4.6.1 Normal ranges:
Normal blood calcium level is between 8.5 to 10.5 mg/dL (2.12 to 2.62 mmol/L) and that of ionized calcium is 4.65 to 5.25 mg/dL (1.16 to 1.31 mmol/L). The amount of total calcium varies with the level of serum albumin, a protein to which calcium is bound. The biologic effect of calcium is determined by the amount of ionized calcium, rather than the total calcium. Ionized calcium does not vary with the albumin level, and therefore it is useful to measure the ionized
calcium level when the serum albumin is not within normal ranges, or when a calcium disorder is suspected despite a normal total calcium level.\(^{(51)}\)

**Corrected calcium level:**

One can derive a corrected calcium level, to allow for the change in total calcium due to the change in albumin-bound calcium. This gives an estimate of what the total calcium level would be if the albumin were a specified normal value. Exact formulae used to derive corrected calcium may depend on the analytical methods used for calcium and albumin. However the traditional method of calculating it is shown below.\(^{(55)}\)

Corrected calcium (mg/dL) = measured total Ca (mg/dL) + 0.8 (4.0 - serum albumin [g/dL]),

where 4.0 represents the average albumin level in g/dL.

In other words, each 1 g/dL decrease of albumin will decrease 0.8 mg/dL in measured serum Ca and thus 0.8 must be added to the measured Calcium to get a corrected Calcium value.

Or: Corrected calcium (mmol/L) = measured total Ca (mmol/L) + 0.02 (40 - serum albumin [g/L]), where 40 represents the average albumin level in g/L.

In other words, each 1 g/L decrease of albumin, will decrease 0.02 mmol/L in measured serum Ca and thus 0.02 must be added to the measured value to take this into account and get a corrected calcium.\(^{(55)}\)

1.3.4.6.2 Hypocalcaemia:

Hypocalcaemia is the presence of low serum calcium levels in the blood. Calcium in the blood exists in three primary states: bound to proteins (mainly albumin), bound to anions such as phosphate and citrate, and as free ionized calcium. Only the ionized calcium is physiologically active. Common causes of hypocalcemia include hypoparathyroidism, vitamin D deficiency, and chronic kidney disease. Symptoms of hypocalcemia include neuromuscular irritability, electrocardiographic changes, and seizures. Treatment is dependent upon the cause, but most commonly includes supplementation of calcium and some form of vitamin D or its analogues.\(^{(55)}\)

1.3.4.6.2.1 Signs and symptoms:

The neuromuscular symptoms of hypocalcemia are caused by a positive bathmotropic effect due to the decreased interaction of calcium with sodium channels. Since calcium blocks sodium channels and inhibits depolarization of nerve and muscle fibers, diminished calcium lowers the threshold for depolarization.\(^{(55)}\)
1.3.6.2.2 Causes:
Hypoparathyroidism is a common cause of hypocalcemia. Calcium is tightly regulated by the Parathyroid hormone (PTH). In response to low calcium levels, PTH induces the kidneys to reabsorb calcium, the kidneys to increase production of calcitriol thereby increasing intestinal absorption of calcium, and the bones to release calcium. These actions lead to a re-balance in the blood calcium levels. However, in the setting of absent, decreased or ineffective PTH hormone, the body loses this regulatory function, and hypocalcemia ensues. Chronic renal failure, Absent active vitamin D, Ineffective active vitamin D, Pseudohypoparathyroidism, Severe acute hyperphosphataemia, Tumourlysis syndrome, Acute renal failure, Exposure to hydrofluoric acid, As a complication of pancreatitis and Alkalosis, caused by respiratory or metabolic alkalosis, the concentration of freely ionized calcium, the biologically active component of blood calcium, decreases. This is because, since a portion of both hydrogen ions and calcium are bound to serum albumin, when blood becomes alkalotic, bound hydrogen ions dissociate from albumin, freeing up the albumin to bind with more calcium, and thereby decreasing the freely ionized portion of total serum calcium. For every 0.1 increase in pH, ionized calcium decreases by about 0.05 mmol/L. This hypocalcemia related to alkalosis is partially responsible for the cerebral vasoconstriction that causes the lightheadedness, fainting, and paraesthesia often seen with hyperventilation.\(^{(56)}\)

1.3.4.6.3 Hypercalcaemia:
Hypercalcaemia is an elevated calcium (Ca\(^{2+}\)) level in the blood. It can be an asymptomatic laboratory finding, but because an elevated calcium level is often indicative of other diseases, a workup should be undertaken if it persists. It can be due to excessive skeletal calcium release, increased intestinal calcium absorption, or decreased renal calcium excretion.\(^{(55)}\)

1.3.4.6.3.1 Signs and symptoms:
The neuromuscular symptoms of hypercalcemia are caused by a negative bathmotropic effect due to the increased interaction of calcium with sodium channels. Since calcium blocks sodium channels and inhibits depolarization of nerve and muscle fibers, increased calcium raises the threshold for depolarization. There is a general mnemonic for remembering the effects of hypercalcaemia: "Stones, Bones, Groans, Thrones and Psychiatric Overtones"
Stones (renal or biliary) Bones (bone pain) Groans (abdominal pain, nausea and vomiting) Thrones (polyuria) Psychiatric overtones (Depression 30-40\%, anxiety, cognitive dysfunction, coma)\(^{(55)}\)
Primary hyperparathyroidism and malignancy account for about 90% of cases of hypercalcaemia, primary hyperparathyroidism, solitary parathyroid adenoma, primary parathyroid hyperplasia, parathyroid carcinoma, multiple endocrine neoplasia (MEN), familial isolated hyperparathyroidism, lithium use, familial hypocalciurichypercalcaemia/familial benign hypercalcaemia, Malignancy, Vitamin-D metabolic disorders, Disorders related to high bone-turnover rates. (56)

A hypercalcaemic crisis is an emergency situation with a severe hypercalcaemia, generally above approximately 14 mg/dL (or 3.5 mmol/l). (57) The main symptoms of hypercalcaemic crisis are oliguria or anuria as well as somnolence or coma after recognition, primary hyperparathyroidism should be proved or excluded. (58) In extreme cases of primary hyperparathyroidism, removal of the parathyroid gland after surgical neck exploration is the only way to avoid death. The diagnostic program should be performed within hours, in parallel with measures to lower serum calcium. Treatment of choice for acutely lowering calcium is extensive hydration and calcitonin, as well as bisphosphonates (which have effect on calcium levels after one or two days. (59)

1.3.4.7 Calcium channel blocker:
Calcium channel blockers (CCB), calcium channel antagonists or calciumantagonistsare a number of medications that disrupts the movement of calcium through calcium channels. Calcium channel blockers are used as antihypertensive drugs, i.e. as medications to decrease blood pressure in patients with hypertension. CCBs are particularly effective against large vessel stiffness, one of the common causes of elevated systolic blood pressure in elderly patients. Calcium channel blockers are also frequently used to alter heart rate, to prevent cerebral vasospasm, and to reduce chest pain caused by angina pectoris.

Despite their effectiveness, CCB's often have a high mortality rate over extended periods of use, and have been known to have multiple side effects. Potential major risks however were mainly found to be associated with short-acting CCBs. Classes into two types: Dihydropyridine and Non-dihydropyridine. (59) calcium channel embedded in a cell membrane. In the body's tissues, the concentration of calcium ions (Ca^{2+}) outside of cells is normally about ten-thousand-fold higher than the concentration inside of cells. Embedded in the membrane of some cells are calcium channels. When these cells receive a certain signal, the channels open, letting calcium rush into the cell. The resulting increase in intracellular calcium has different effects in different
types of cells. Calcium channel blockers prevent or reduce the opening of these channels and thereby reduce these effects.

There are several types of calcium channels, and a number of classes of calcium channel blockers, but almost all of them preferentially or exclusively block the L-type voltage-gated calcium channel. \(^{(59)}\)

1.3.4.8 Calcium and Hypertension:

Calcium is a major mineral essential for human health. In addition to playing an important role in building and maintaining strong bones and teeth, calcium aids in muscle contraction and regulates blood pressure. In fact, consuming a diet rich in calcium, along with other minerals, such as potassium and magnesium, is recommended for reducing blood pressure.

Calcium may reduce blood pressure because it plays a vital role as a mediator in the constriction and relaxation of blood vessels, according to the Linus Pauling Institute at Oregon State University. Similarly, calcium is essential for muscle contraction and relaxation. Consuming a calcium-deficient diet causes a concentration of calcium ions in the intercellular fluid. This elevates both the active form of vitamin D, vitamin D3, and the parathyroid hormone resulting in an increased calcium concentration in smooth muscle cells. This shift in calcium concentration increases vascular resistance, raising blood pressure according to an article published in the Journal of the American College of Nutrition in February 2009.

1.3.5. Essential arterial hypertension and stone disease:

A significant percentage of hypertensive subjects has a greater risk of renal stone formation, especially when hypertension is associated with excessive body weight. Higher oxaluria and calciuria as well as supersaturation of calcium oxalate and uric acid appear to be the most important factors. Excessive weight and consumption of salt and animal proteins may also play an important role. \(^{(59)}\)
1.3.6. Previous studies:

1.3.6.1 Previous study of uric acid:

Plasma UA level was not associated with incident hypertension in older men; the association that was observed among men who were younger than 60 yr was confounded in fully adjusted models. In University of Washington Medical Center. Our study is the first to examine this association in older men, who shoulder the larger share of the hypertension disease burden ($p \leq 0.001$). This study also is the first to control simultaneously for renal function and metabolic factors, including insulin resistance and dyslipidemia, in addition to other confounders. Our findings should be confirmed by subsequent studies in older individuals; moreover, future investigations of the UA–hypertension relation should control for metabolic factors that may confound this association.\(^{(60)}\)

1.3.6.2 Previous study of calcium:

Calcium plays an important role in the pathophysiology of essential hypertension. Serum calcium levels were measured in 117 subjects with essential hypertension and 77 first-degree relatives. The results showed that serum calcium levels were significantly ($p<0.01$) decreased in both males and females with essential hypertension and their first-degree relatives when compared with the normotensive controls. This is the first study in Indian population.\(^{(61)}\)
1.4 Objectives:

1.4.1 General objective:

To assess the plasma Uric acid and calcium among Sudanese hypertensive patients.

1.4.2 Specific objectives:

1- To compare plasma uric acid and calcium levels between tests and controls.
2- To correlate between plasma uric acid and calcium among duration of disease.
Chapter two

2. Materials and methods

2.1 Materials:

2.1.1 Study design:-
This is a quantitative, descriptive, analytic, cross-sectional and hospital-based study.

2.1.2 Study area and period:-
This study was conducted in Khartoum state, capital and central of Sudan country. Patients enrolled in this study were come to refer clinic of Aldosogi specialized hospital, during the period from March 2014 to July 2014.

2.1.3 Study population:-
Study included Sudanese hypertensive males and females above 40 years old.

2.1.4 Sample size:-
Population of this study was categorized into a study group of 60 hypertensive patients and a control group of 40 healthy subjects (non hypertensive).

2.1.5 Inclusion criteria:-
Sudanese patients with hypertension where included as a test group and healthy subjects as control.

2.1.6 Exclusion criteria:-
Patients with renal disease, gout, osteomalacia, Paget’s disease and parathyroid disorder.

2.1.7 Ethical consideration:-
Firstly the permission of this study was obtained from medical directors of, then all participants were told about the research aims and benefits during interview and all of them were agreed to participate in addition to all samples were taken from participants after their agreement and filling the questionnaire.

2.1.8 Data collection and clinical examination:
Clinical data for every patient was collected by questionnaire using refer card for patient while clinical examinations done by clinicians in above mentioned hospital.
2.1.8.1 Sampling:
Blood sample “3ml” were from subjects of study group after fulfillments of questionnaire as well as control group, using disposable syringe and sprit for sterilization the area of collection. Collected blood was drawn in heparin containers and gently mixed with anticoagulant. The sample in heparin containers was separated by centrifuged it at 3000 rpm for 5 min. Hemolized and lipamic samples were rejected and excluded from the study. Obtained plasma was tested for uric acid using BiosystemBTS-310.

2.2 Methods:
2.2.1 Measurement of uric acid:
URICASE/PEROXIDASE method, enzymatic, colorimetric method.

2.2.1.1 Principle:
The assay is based on the oxidized uric acid by uricase to give hydrogen peroxide and allantoin, the fast react with 4-aminoantipyrine and phenol to give pink color which can measure at 520nm.

\[
\text{Uric acid} + O_2 \xrightarrow{\text{uricase}} \text{Allantoin} + CO_2 + H_2O_2
\]

\[
2H_2O_2 + 4\text{–Aminoantipyrine} + \text{phenol} \xrightarrow{\text{peroxidase}} \text{Quinoneimine} + 4H_2O
\]

2.2.1.2 Procedure:
The sample in heparin container was separated by centrifuged it at 3000 rpm for 5 min and the test was done as follow

1-Assay conditions:

Wavelength……………………………..520nm

Cuvette………………………………1 cm. light path

Temperature…………………………..37°C /15-25°C

2-The instrument was Adjusted to zero with distilled water

3-Pepette into cuvette:
<table>
<thead>
<tr>
<th></th>
<th>Standard</th>
<th>Sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>WR(ml)</td>
<td>1.0</td>
<td>1.0</td>
</tr>
<tr>
<td>Standard(µl)</td>
<td>25</td>
<td>—------</td>
</tr>
<tr>
<td>sample(µl)</td>
<td>—</td>
<td>25</td>
</tr>
</tbody>
</table>

4- Mix thoroughly and incubate the tubes for 10 minutes at room temperature or for 5 minutes at 37°C.

5- Measure the absorbance at 520 nm against the blank.

**2.2.1.3 Calculation:**

\[
	ext{Uric acid (mg/dl)} = \frac{\Delta \text{Absorb}}{\Delta \text{Absorb std}} \times \text{con of std} (6 \text{ mg/dl})
\]

Conversion factor: mg/dl *357= µmol/L.

**2.2.1.4 Reference range:**

**Serum or plasma:**

- Men 3.5-7.2 mg/dl
- Women 2.6 – 6.0 mg/dl\(^{(30)}\)

**2.2.2 Measurement of calcium:**

CALCIUM – MTB (METHYLTHYMOL BLUE) method, enzymatic, colorimetric method.

**2.2.2.1 Principle:**

The assay is based on the reaction of calcium with methylthymol blue (MTB) in alkaline medium to give complex color which can measure at 600 nm.

**2.2.2.2 Procedure:**

1- **Sample Precaution:**

- Serum or heparinized plasma.
- Sample without hemolysis.
- Position of patient horizontal position and relaxes state.
- Calcium effect by diet taken of fasting state.
- Calcium highly contaminated by (glass, ware, tube) so wash the material by 5% HCL.
- Do not use tourniquet.
The sample in heparin container was separated by centrifuged it at 3000 rpm for 5 min and the test was done as follow

2-Assay conditions:
Wavelength………………………………610nm.
Cuvette……………………………………1 cm. light path.

Temperature………………………………37°C /15-25°C

2- The instrument was Adjusted to zero with distilled water.

3-Pepette into cuvette:

<table>
<thead>
<tr>
<th></th>
<th>Standard</th>
<th>Sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>WR(ml)</td>
<td>1.0</td>
<td>1.0</td>
</tr>
<tr>
<td>Standard(µl)</td>
<td>10</td>
<td>—</td>
</tr>
<tr>
<td>sample(µl)</td>
<td>—</td>
<td>10</td>
</tr>
</tbody>
</table>

4- Mix thoroughly and incubate the tubes for 2 minutes at room temperature.

5- Measure the absorbance at 610 nm against the blank.

2.2.2.3 Calculation:

\[
\text{Calcium (mg/dl)} = \left( \frac{\Delta \text{Abs} \text{fT}}{\Delta \text{Abs} \text{fstd}} \right) \times \text{conof std} (10 \text{ mg/dl})
\]

Conversion factor: mg/dl *0.25= µmol/L.

2.2.2.4 Reference range:
- Serum or plasma: 8.6 – 10.3 mg/dl^{(51)}

2.2.3 Quality Control:-

The precision and accuracy of all methods used in this study were checked each time a batch was analyzed by including commercially prepared control sera.

2.2.4 Statistical analyses :-

Data was analyzed by independent t-test and bivariate correlation by the programmed computer (SPSS).
CHAPTER THREE

RESULT

3. Result:
This study was conducted on 60 patients with hypertension as test group and 40 control groups (healthy volunteers). Age and gender of the test group was match with control group. In the present study, male account 70% (n=42) from the test and 67.5% (n=27) from control group, while female account 30% (n=18) from the test and 32.5% (n=13) from control group.

Table (3-1) showed significant difference between the mean of calcium in mg/dl in study group patients and control group. (8.2±0.46mg/dl) versus (8.8±0.44) (P=0.049 significant at ≤0.05) and also insignificant difference between the mean of uric acid in mg/dl in study group patients and control group. (5.4±0.90 mg/dl) versus (5.2±0.83 mg/dl) (P=0.126 insignificant at >0.05).

Figure (3-1) A scatter plot shows the weak negative correlation between calcium and duration of hypertension (R = -0.3, P=0.049).

Figure (3-2) A scatter plot shows the weak positive correlation between uric acid and duration of hypertension (R = 0.2, P=0.126).
**Table (3-1):**

Show the mean, standard deviation and probability in calcium and uric acid (mg/dl) in hypertensive patients and non-hypertensive.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Test group (HTN) (n=60)</th>
<th>Control group(non-HTN) (n=40)</th>
<th>P. value</th>
</tr>
</thead>
<tbody>
<tr>
<td>calcium (mg/dl)</td>
<td>8.2± 0.46</td>
<td>8.80±0.44</td>
<td><strong>0.049</strong></td>
</tr>
<tr>
<td>Uric acid (mg/dl)</td>
<td>5.4± 0.90</td>
<td>5.2±0.83</td>
<td><strong>0.126</strong></td>
</tr>
</tbody>
</table>

- The table shows the mean± Std. deviation and probability (P).
- Independent t- test was used for comparison.
- P- Value ≤ 0.05 was considered significant.
**Fig (3-2):-**

A scatter plot show the relationship between uric acid (mg/dl) and duration of hypertension (years), \( r=0.09, P=0.126 \).
Fig (3-4):

A scatter plot shows the relationship between calcium (mg/dl) and duration of hypertension (years), ($r = -0.179, P=0.049$).
CHAPTER FOUR
Discussion, Conclusion and Recommendations

4.1Discussion:
Hypertension is one of the most common worldwide diseases afflicting humans and is a major risk factor for stroke, myocardial infarction, vascular disease, and chronic kidney disease. Calcium aids in muscle contraction and regulates blood pressure. In fact, consuming a diet rich in calcium, along with other minerals, such as potassium and magnesium, is recommended for reducing blood pressure. Calcium may reduce blood pressure because it plays a vital role as a mediator in the constriction and relaxation of blood vessels, according to the Linus Pauling Institute at Oregon State University. Similarly, calcium is essential for muscle contraction and relaxation. Consuming a calcium-deficient diet causes a concentration of calcium ions in the intercellular fluid. This elevates both the active form of vitamin D, vitamin D3, and the parathyroid hormone resulting in an increased calcium concentration in smooth muscle cells. This shift in calcium increases vascular resistance, raising blood pressure according to an article published in the Journal of the American College of Nutrition in February (2009).

In this study there was significant difference in the mean of calcium level of the test group when compared with control group significant (P= 0.049), this result agree with the result observed by (K. Sudhakar, M. Sujatha and S. Ramesh Babu) whom reported that there was significant difference.

The present data demonstrated that there is a significant weak negative correlation between calcium and duration of hypertension disease (r= - 0.179, P=0.049), this result agree with the result observed by (K. Sudhakar, M. Sujatha and S. Ramesh Babu) whom said that there was slightly lower relation between the period of hypertension and plasma calcium result from the effect of the regulations that control calcium.

Uric acid was first associated with primary hypertension in 1874, yet its role in this condition remains unclear. Historically, uric acid was thought to be a secondary response to hypertension or its associated conditions. However, more recent experimental suggest that uric acid could have a contributory role in the pathogenesis of elevated blood pressure. More studies are needed to help dissect the potential mechanisms by which uric acid could initiate this response. It
remains possible that uric acid is a marker for xanthine oxidase-associated oxidants and that the latter could be driving the hypertensive response. However, the weight of the evidence suggests that uric acid is a true modifying and possibly causal factor for human primary hypertension. Hence, early management of hyperuricemia might delay the development of essential hypertension.

Also, in this study, there was no significant difference in the mean of uric acid level of the test group when compared with control groups (significant, P = 0.126), this result disagree with the result observed by (University of Washington Medical Center (M.M., J.H., Y.-G.K., J.A.J., D.-H.K., K.L.G., R.J.J.), Seattle; the Division of Nephrology) whom reported that there was significant difference in serum uric acid (p ≤ 0.001).

The present data demonstrated that there is a significant weak positive correlation between uric acid (mg/dl) and duration of hypertension disease (r = 0.09, P = 0.126), this result agree with the result observed by ((M.M., et al), Seattle; the Division of Nephrology) whom said that there was weak increase relation between the period of hypertension and plasma uric acid.

Most of hypertensive subjects has a greater risk of renal stone formation, especially when hypertension is associated with excessive body weight. Higher oxaluria and calciuria as well as super saturation of calcium oxalate and uric acid appear to be the most important factors.

Excessive weight and consumption of salt and animal proteins may also play an important role (Loris Borghi, Istituto di Semeiotica Medica, Università degli Studi di Parma, Via Gramsci, 14, 43100 Parma, Italy).
4.2 Conclusion:

- Calcium plasma levels are significantly increased in hypertensive patient’s compared to control.
- Plasma levels of uric acid are insignificantly increased in hypertensive patients compared to control.
- There is an insignificant positive correlation between uric acid and duration of hypertension disease.
- There is a significant negative correlation between calcium and duration of hypertension disease.
4.3 Recommendations:

From the results of this study, it is recommended that:

- Renal function tests should be checked regularly in hypertensive patients, especially in those with a prolonged period of the disease.
- Health education, diet control, and exercise are important factors to achieve good control of hypertension.
- Measuring of hormones regulating calcium homeostasis.
- Ionized calcium level useful to measure when the serum albumin is not within normal ranges, or when a calcium disorder is suspected despite a normal total calcium level.
References:


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K. Sudhakar, M. Sujatha and S. Ramesh BabuSerum calcium levels in patients with essential hypertension and their first degree relatives.
Sudan University of Sciences and Technology
College of Graduate Studies

Question of:
Assessment of the plasma calcium and uric acid in Sudanese hypertension

General information:
1. Name ................................................................. No ( )
2. Age ................................. years
3. Sex Male ( ) Female ( )
4. Duration of disease ..................... years

Laboratory diagnosis:

<table>
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<tr>
<th>Investigation</th>
<th>Result</th>
<th>Reference</th>
<th>Duration</th>
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<td>Calcium</td>
<td>8.5 - 10.5 mg/dL</td>
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<tr>
<td>Uric acid</td>
<td>3.5 - 7.2 mg/dL</td>
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