Measurement of Calcium, Inorganic Phosphate and Albumin Levels in Serum of Iraqi Hypertensive Male Patients

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Abstract

Hypertension is a major health problem in adults, and contributes to cardiovascular disease. Malnutrition or deficiency in different nutrients found to be a risk factor for pathogenesis of essential hypertension. No clear association of calcium with blood pressure level could be identified. In this work serum calcium and albumin in hypertensive patients were compared with healthy control. Thirty uncomplicated, untreated, hypertensive, but otherwise have no other systemic diseases males, ranging in age from (26–55) years old, were entered into the study. Their blood pressure values were more than 95/140 mm Hg (seated posture). Control group consists of thirty healthy males with normal blood pressure and their age range is 22-46 years old. Serum calcium and serum albumin were measured calorimetrically using ready for use kits. Results showed a significant decrease ($p<0.05$) in serum calcium of patients group as compared with control group. No significant differences noticed between the two groups in serum albumin ($p>0.05$). Correlation coefficients between serum albumin and calcium in control group and patients group are 0.13, and 0.12, respectively. The ratio between mean serum albumin on mean serum calcium showed an increase in patients group (23.387) in comparing with control group (19.348). In this research we conclude that hypertensive patients have significantly lower serum total calcium than control group. Calcium supplements may be required as adjuvant treatment in addition to decrease NaCl in the diet to reduce excretion of calcium in urine. There is no significant difference between the two groups in serum albumin. In this work a new ratio (S. albumin/ S. calcium) is introduced as a useful marker for calcium state. No correlation was found between serum albumin and calcium in both groups indicating the need for estimating the free calcium in both groups. Further studies required for other biochemical parameters in larger patients sample size.

Keywords: hypertension, albumin, calcium, blood pressure.

قياس مستويات الكالسيوم والفوسفات اللاعضوي و الالبومين في امصال ذكور المرضى العراقيين المصابين بارتفاع ضغط الدم

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الخلاصة:

ان ارتفاع ضغط الدم من المشاكل الصحية الرئيسية عند البالغين ويساهم في التسبب بأمراض القلب والشرايين. إن سوء التغذية ونقص بعض المواد الغذائية قد تكون من مسببات هذا المرض. لا توجد علاقة واضحة بين مستوى الكالسيوم في الدم ومرض ضغط الدم. في هذا البحث تم قياس تراكيز الكالسيوم والألبومين عند مرضى ضغط الدم ومقارنته بمستوياتهما عند الناس الأصحاء. لقد اشترك في هذه الدراسة 30 شخصا (ذكور فقط) مصابا بمرض ضغط الدم فقط وليس لديهم أمراض أخرى أعمارهم بين 26 و55 سنة. كان مستوى ضغط الدم لدى المرضى أعلى من 95/140 ملليمتر زئبق مقاسا بوضعية الجلوس. اخذ 30 شخصا سليما من المرض لغرض المقارنة أعمارهم بين 21 و44 سنة. كل المرضى لم تتم معالجتهم بأي دواء. تم قياس تركيزي الكالسيوم والألبومين بالطرق اللونية باستعمال العدد القياسية الجاهزة. أظهرت النتائج انخفاضا معنويا ($p<0.05$) في تركيز الكالسيوم في أمصال مرضى ضغط الدم مقارنة بمجموعة السيطرة بينما لا يوجد اختلاف معنويا ($p>0.05$) في تركيز الالبومين بين المجموعتين. كان معامل الارتباط بين تراكيز الكالسيوم والألبومين منخفضا لمجموعة المرضى ومجموعة السيطرة وبلغ 0.13 و0.12 ، على التوالي. ان قيمة نسبة معدل تراكيز الألبومين الى الكالسيوم (ألبومين/كالسيوم) هي نسبة عالية لدى مجموعة المرضى وتبلغ 23.387 مقارنة بمجموعة السيطرة 19.348. نستنتج من هذا البحث بان مرضى ضغط الدم لديهم تركيز اقل من الكالسيوم في المصل مقارنة بمجموعة السيطرة. لا يوجد اختلاف معنويا في تركيز الألبومين بين المجموعتين. وفي هذا البحث استحدثت نسبة جديدة هي نسبة معدل الألبومين الى معدل الكالسيوم في المصل كدليل مفيد لحالة الكالسيوم في الجسم. لا يوجد ارتباط بين تراكيز الألبومين والكالسيوم في مصول كلا المجموعتين مشيرا الى أهمية تقدير الكالسيوم الحر لدى المرضى بالإضافة الى الكالسيوم الكلي. يوصى بإضافة الكالسيوم كمساند الى برنامج علاج المرضى وتقليل تناول ملح الطعام لتقليل طرح الكالسيوم في الإدرار ودراسة مواد كيموحيوية أخرى لدى مرضى ارتفاع الضغط وبحجم عينات اكبر.

الكلمات الدالة: ارتفاع ضغط الدم، الألبومين، الكالسيوم، ضغط الدم

Introduction

Hypertension was defined as blood pressure greater than 140/90 mmHg. High blood pressure results from either an increased output of blood by the heart or, most often, increased resistance to blood flow in the arteries. In those with high blood pressure, the heart must work harder than normal to force blood through the arteries. Hypertension is a major health problem in adults, and contributes to cardiovascular disease. Hypertension, the most common cardiovascular disease, affect about 30% of Americans over age 50 and 50% by age 74 and only about 27% of American hypertensive have their blood pressure under control ,prevalence of hypertension is showing alarmingly steep rise due to rapid changes in diet and lifestyle. There are two types of hypertension; primary (essential hypertension) which accounts for 90%of cases and secondary hypertension which account for 10% of cases is secondary from other identifiable disorders ,about half of hypertensive are 'salt-sensitive', meaning that their blood pressure will decrease significantly when salt intake is restricted [1,2].

Positive family history of cardiovascular diseases and other atherosclerosis risk factors, obesity, high salt intake are known to be a major risk factor in hypertension, hypercholesteremia is a risk factor for atherosclerosis and subsequent hypertension [3].

Malnutrition or deficiency in different nutrients found to be a risk factor for pathogenesis of essential hypertension , such as zinc , vitamin C and deficiency in different other antioxidants [4].

There are many repots about alteration in the trace elements and other metals level and hypertension. Altered plasma status of copper, zinc, magnesium and calcium in hypertension has been reported [5]. Dietary intakes of potassium and magnesium have been reported to have favorable effects on blood pressure. Intake of these elements and calcium reduce blood pressure [6]. Cholesterol and nicotine are also risk factors for hypertension [7].

Current American guidelines recommend weight control, reduced intake of sodium chloride (salt), reduced alcohol consumption, and possibly increased dietary potassium as nutritional approaches to prevent and treat hypertension [8].

A relationship of calcium and magnesium intake with BP levels in the young has been studied, with variable

results. No clear association of potassium or calcium with BP level could be identified an Investigational effort to detect an independent effect of a dietary Action on BP level is complicated by the intercorrelation of multiple nutrients in the diet. A possible relationship between increase serum level of different substances including vitamin D and calcium, and cardiovascular disease was recorded [9,10].

In observational studies, significant inverse associations of blood pressure with intake of magnesium, potassium, calcium, fiber, and protein have also been reported [11]. However, in trials that tested these nutrients, often as dietary supplements, the reduction in blood pressure has typically been small and inconsistent [(12)]. Hence, in this study, serum calcium, inorganic phosphate, and albumin in hypertensive patients were compared with healthy control. In addition, serum albumin/serum calcium ratio was tested as indicator for calcium state in blood. The aim is obtaining a recommendation about using calcium supplements as a mean for treatment by altering serum calcium (if present).

Materials and Methods

Thirty uncomplicated, untreated, hypertensive, but otherwise have no other systemic diseases males were entered into the study. Their age expressed as mean ± standard deviation was (54.7±11.5) years old. Each hypertensive subject had a blood pressure measurement by conventional sphygmomanometry in excess of 90/140 mm Hg (seated posture), with the arm in the horizontal position after 5 min of quiet sitting, and had never received any antihypertensive treatment. The study was performed under out-patient conditions. Control group consists of thirty healthy males with normal blood pressure and their age range is between 22 and 46 years old. When the heart contracts (systole), blood pressure increases; when the heart relaxes (diastole), the pressure decreases. Average blood pressure reading for young adults in good physical condition are 110-120mmHg systolic over 70-80mmHg diastolic.

Five milliliters of venous blood samples were collected from cases without tourniquet. Sera were separated and measured immediately or, if necessary, stored at (-20° C) until analysis. Serum calcium measured by O-cresolphthalin complexion method [13] and serum albumin by bromocresol green method [14] using the procedures described by Randox® kits leaflets. The inorganic phosphorous was estimated in serum

using BioMerieux® kit. The method is colorimetric determination without deproteinization using a single reagent (ammonium hepta-molybdate) which reacts with serum phosphorous to form a phosphomolybdate complex in the presence of a reducing agent (ferrous sulfate). The colored complex is measured at 690 nm.

Results

The results of serum calcium and albumin in patients and control groups expressed as (Mean ± Standard deviation) in addition to the S.Albumin /S.Ca^{+2} ratios are represented in Table 1.

Serum calcium concentration was decreased in patients when compared with healthy control (1.84± 0.44, 2.27±0.22 mmol/L respectively) with P- value < 0.05.

The same result obtained when compare the level of serum albumin, there was decrease in this level in hypertensive patients when compared with healthy control, but this decrease was statistically not significant (P-value> 0.05), (41.55±7.72,44.76±5.74 g/ L respectively) .

The ratio between mean serum albumin on mean serum calcium showed an increase in patients group (23.387g/mmol) in comparing with control group (19.348 g/mmol).

Regarding the serum inorganic phosphate concentration, there was decrease in patients as compared with control (0.98±0.45, 1.17±0.14 mmol/L respectively) ,but this difference is not significant P- value > 0.05 .

Discussion

The decrease in serum calcium noticed in the present work is in accordance with other researches. In one research both serum and intralymphocytic concentrations of Na^{+}, and Ca^{+2} in hypertensive group were significantly higher than those in the normotensives [12]. Several experimental and clinical studies suggest that calcium depletion elevates blood pressure. Also the results support the studies related to the treatment of hypertension by mineral supplements including calcium [15].

Calcium load leads to the increment in Na excretion [16] and a reduced sodium intake reduced calcium excretion and *vice versa* and hence decreases in hypertension. The maintained natriuretic ability found in patients with higher salt sensitivity

could be mediated not only by the degree of underlying volume expansion but also by a well-known natriuretic effect of calcium (Ca^{+2}) through an increase in the renal tubular Ca^{+2} concentration [17].

Table 1. omparison between Calcium, Inorganic Phosphate and Albumin levels in serum of Iraqi hypertensive male patients and control group,represent as mean± SD.

	Patients (n=30)	Healthy Control (n=30)	P-value
S.Calcium(mmol/L)	1.84± 0.44	2.27 ± 0.22	2.063E-05
S.Albumin(g/L)	41.55 ±7.72	44.76 ± 5.74	0.09
S.Phosphate(mmol/L)	0.98 ± 0.45	1.17 ± 0.41	0.084
S.Albumin / S.Ca++(g/mmol)	0.98 ± 0.45	1.17 ± 0.41	

These changes in the blood pressure with low serum calcium level could possibly be attributed to some poorly known alterations in cell membrane transport mechanisms. Abnormal cellular ion transport resulting in altered membrane control over intracellular calcium may be related to essential hypertension [18]. The free intracellular calcium concentration determines the tension in vascular smooth muscle cells, thereby resulting in peripheral vascular resistance. Calcium has direct effect on peripheral vascular tone. Alternations in intracellular calcium are thought to be involved in the common pathway mediating the secretion and action of many hormones, including the presser action of catecholamine and angiotensin II [19]. Ionized serum Ca is reported to be lower in low-renin hypertensive patients and higher in high-renin hypertensive patients than in normal-renin hypertensive or in normotensives. Plasma renin activity in essential hypertension has a continuous negative correlation with serum Mg and a positive correlation with serum ionized Ca. Hence, plasma renin in hypertension may reflect (or contribute to) Ca and Mg flux changes across cell membranes [20].

Another notable factor which is indirectly involved in the pathogenesis of essential hypertension is altered lipid metabolism in the situation of low serum calcium level or decreased dietary calcium intake. Low calcium diet or low serum calcium stimulates increased production of 1,25-dihydroxyvitamin D which in turn,

stimulates adipocyte Ca2+ influx and, as a consequence, stimulates lipogenesis, suppresses lipolysis, and increases lipid accumulation; whereas increasing dietary calcium inhibits these effects and markedly accelerates fat loss. Many researchers even recommend a regular consumption of the recommended daily levels of dietary calcium to combat with hypertensive disorders [21]. Jolma P et al. [22] found that calcium supplementation reduced blood pressure in hypertensive individuals during chronic nitric oxide synthase inhibition and abrogated the associated impairments in endothelium-dependent and endothelium-independent arterial relaxation. High calcium diet had been found to enhance the vasorelaxation in nitric oxide-deficient hypertension.

The research observations may elucidate the mechanism(s) by which oral Ca^{+2} supplementation decreases blood pressure in patients with salt-sensitive hypertension [23]. Significant inverse correlation of blood pressure with the increase intake of some metals including calcium and different other nutrients have been frequently recorded. More recently the Dietary Approach to Stop Hypertension (DASH) [24] clinical trial in adults with high, normal, and untreated stage I hypertension demonstrated that significant BP reduction occurred on a diet high in fruits and vegetables and low-fat dairy products. Diets with high fiber content of, which may have impeded calcium absorption [25] while there is also some evidence that other diet nutrients, including potassium, calcium, and magnesium, are inversely related to BP levels, the BP level is higher in those with diets lower in potassium, calcium, magnesium, and vitamins. These results suggest that diets deficient in multiple nutrients may contribute to the development of hypertension in adolescents having risk factors for cardiovascular disease. These observations are consistent with the dietary benefits on BP level observed in the DASH trial [24]. Indeed, several studies suggest that the effect of Na intake on blood pressure is determined by the adequacy of other minerals, such as Ca^{+2} , Mg^{+2} and K^{+} . [26].

The presser effect of NaCl seems to be expressed in subjects with the lowest intake of these minerals [21]. The natriuretic effect of Ca^{+2} may be mediated either through increases in serum and/or renal tubular Ca^{+2} concentrations. In one research, oral intake of calcium lactate tablet increased Urinary excretion of calcium and could reduce their average blood pressure and prevent them from

hypertension for those with mild and moderate hypertension and in high risk with higher blood pressure [27].

The results of this research revealed additional evidence to the fact that the retention in calcium excretion and subsequent maintenance in serum calcium is good for hypertension.

The kidney plays a key role in the maintenance of mineral ion homeostasis, particularly that of calcium (Ca^{+2}). Urinary calcium (UCa^{+2}) excretion depends on the filtered load of Ca^{+2} and on several other factors, including PTH levels, sodium (Na^{+}) excretion, serum concentrations of Ca^{+2} and magnesium (Mg^{+2}), Ca^{+2} intake and absorption, and acid-base status [28]. The effect of NaCl loading is hypercalciuric and hence decreases in serum calcium. In those who are salt sensitive and in whom dietary calcium is suboptimal dietary replenishment of calcium may reduce blood pressure. Volume expansion with saline causes a decline in Na^{+} and Ca^{+2} reabsorption by the kidney proximally and distally, thereby resulting in an increase in the excretion of both cations. Untreated hypertensive patients had a higher prevalence of hypercalciuria in patients with essential hypertension. It was concluded that hypercalciuria is a frequent finding of untreated essential hypertension [29].

Hypertensive subjects have been shown to present an increase in urinary calcium excretion despite a lower dietary calcium intake ; this apparent paradox has been referred to as a renal calcium leak, and several studies have clearly suggested that urinary calcium excretion was significantly higher in salt-sensitive subjects compared with those who were salt-resistant [15].

The results also indicate that there is no significant difference in the serum inorganic phosphate in patients and control groups. Hence the change in calcium may not dependent on the changes in serum inorganic phosphate only but may involve the free ionic calcium in addition to the bound calcium. In one hypertension research unit, the plasma Aldosterone concentration and parathyroid hormone are high. At the same time we found calcium metabolism alterations: high urine calcium excretion, low serum ionic calcium [30]. These alterations may explain the indifference in serum albumin and decrease in serum calcium in hypertensive patients noticed in our research. In only one study proteinuria (macroalbuminuria) was found to correlate positively and very significantly with both systolic and diastolic blood pressure, but this study needs more confirmation because we didn’t find any corresponding results.

Serum albumin may be affected if there is renal damage [31] and our patients group has no renal complications as mentioned in the subjects and methods paragraph, while another study revealed a positive association was found between serum albumin and blood pressure, since albumin in contrast to high blood pressure is consider to be cardio protective, probably effect cardiovascular risk by unrelated mechanisms.

In the present work, the ratio between serum albumin and serum calcium ratio may be more useful indicator for any change in calcium even it has not noticed statistically. The ratio need more studies in different other diseases related to calcium metabolism to knowledge evaluate its advantages as predictor for these diseases.

Conclusion

Hypertensive patients have significantly lower serum total calcium than control group. There is no significant difference between the two groups in serum albumin and inorganic phosphate. In this work, a new ratio (S.albumin/ S. calcium) is introduced as a useful marker for calcium state. There is a need for estimating the free calcium in both groups. Calcium supplements may be required as adjuvant treatment in addition to decrease NaCl in the diet to reduce excretion of calcium in urine. Further studies required for other biochemical parameters in larger patients sample size.

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