Iron Status in Patients with Primary Hypertension

By

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Abstract:

Hypertension is a major health problem in adults, and contributes to cardiovascular disease. Iron represents a paradox for human health by being essential for many important biological process, but also having an ability to be harmful in many different. In the present study, an attempt is carried out to define the iron status in Iraqi hypertensive patients. The aim is to obtain a recommendation about the risk of increased iron indices as a marker for subsequent ischaemic heart diseases.

Eighty-eight hypertensive uncomplicated, hypertensive patient, but otherwise have no other systemic diseases patients aged 49±13 years old, were entered into the study. Serum levels of iron,, total Iron Binding Capacity (TIBC), and ferritin were measured while Unsaturated iron-binding capacity (UIBC), estimated total iron body stores (ETIBS), transferrrin saturation percentage (TS%) and transferrin concentration were calculated mathematically.

The results showed a significant difference ($p<0.05$) in all iron indices of hypertensive patients in comparing with healthy control group except TIBC, UIBC, and transferrin concentrations, which decrease in these patients in comparing with control group. There is a significant increase in serum ferritin and EIBS in male group as compared with female group. While all other iron indices were differ insignificantly between groups.It can be concluded that hypertensive patients have significantly higher level of iron parameters than control group. There is no significant difference between male and female groups in most iron indices except in ferritin and subsequently EIBS levels. Further studies required for other biochemical parameters in larger patients sample size.

Keywords: *Iron, TIBC, Ferritin, and Hypertension.*

Introduction

High blood pressure (BP) 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(Fahey T.D., Insel P.M.). Hypertension is a major health problem in adults, and contributes to cardiovascular diseases (The Sixth Report of the Joint National Committee). Prevalence of hypertension is showing alarmingly steep rise due to rapid changes in diet and lifestyle (Saladin K.S., Porth C.M. (1998). 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 [,Saladin K.S., Porth C.M. (1998]. In Iraq, the prevalence of diagnosed hypertension in 2004 is 4,664,465 out of 25,374,691 people, which represents 18.4% of the Iraqi population. **While undiagnosed hypertension is estimated to be** 1,399,339 out of 25,374,691 which represents 5.5% of the Iraqi population [**US Census Bureau].**

Investigational efforts to detect an independent effect of a dietary cation on blood pressure level are complicated by the intercorrelation of multiple nutrients in the diet [Ascherio A, Rimm]. Iron represents a paradox for human health by being essential for many important biological process, but also having an ability to be harmful in many different [McCord J. Iron, PremPonka]. Iron plays important roles in many important processes in life, serving as metal cofactor for many enzymes (oxidases, peroxidase, catalases, mitochondrial aconitase, ribonucleotide reductase etc.), and as a component of hemoproteins (hemoglobins, cytochromes). On the other hand, chemical properties of iron can cause damage to the biological systems. At physiological pH and oxygen tension, Fe(II) is readily oxidized to Fe(III), which rapidly forms essentially insoluble $Fe(OH)_3$ polymers and plays a key role in the formation of harmful oxygen radicals that ultimately cause peroxidative damage to vital cell structures [McCord J. Iron, Halliwell B & Gutteridge]. Thus, maintain of iron level within the useful and harmless level is very important because the decrease in iron level may anemia and high level causes damage to the tissues.

In the present study, an attempt is carried out to define the iron status in Iraqi hypertensive patients. The aim is to obtain a recommendation about the risk of increased iron indices as a marker for subsequent ischaemic heart diseases.

Subjects and Methods

Patients: After oral consent, 88 hypertensive uncomplicated, hypertensive patient, but otherwise have no other systemic diseases patients aged 49±13 years old, were entered into the study. Each hypertensive subject had a blood pressure measurement by conventional sphygmomanometry in excess of 95/140 mmHg (seated posture), with the arm in the horizontal position after five minutes of quiet sitting. The study was performed under out-patient conditions in Al-Furat Al-Ausat hospital in Najaf governorate. Control group consists of thirty healthy persons with normal blood pressure and their age range is comparable with patients group. Exclusion criteria included a history of infection, inflammation, cancer, diabetes mellitus, and congestive heart failure.

Measurements: Blood samples were collected from individuals in the morning plain tubes for serum separation by centrifugation in order to estimate the iron status parameters and hormones level. Serum levels of iron were estimated using Ferrozine colorimetric method [Artiss, J.D., Vinogradov], total Iron Binding Capacity (TIBC) were estimated colorimetrically by the following procedure [International Committee]: An excess of iron is added to the serum iron to saturate the transferrin. The unbound iron is precipitated with basic magnesium carbonate. After centrifugation, the iron in the supernatant was determined.Unsaturated iron-binding capacity (UIBC), the amount of protein (apotransferrin) still available to bind iron, can be estimated from the formula: UIBC=TIBC – Serum iron.

The ferritin quantitative kit based on a solid phase enzyme-linked immunosorbent assay (ELISA) was supplied by Monobind® Inc. USA. The assay system utilizes one rabbit anti-ferritin antibody for solid phase (microtitre wells) immobilization and a mouse monoclonal anti-ferritin antibody in the antibody-enzyme horseradish peroxidase (HRP) conjugate solution.

Estimated Total Iron Body Stores (ETIBS) were calculated using the following formula (Rayssiguier, Y.,): *ETIBS (in µmol) = (serum ferritin in µg/L) * 143*

Transferrrin saturation percentage (TS%) was calculated from the following equation (Freeman V. and Arneson):

*TS%=(Serum Iron/TIBC)*100%*

Transferrin concentration can be calculated using the following formula (Morgan EH)

*Transferrin Conc. (g/L) =S.Iron (µmol/L)/ (TS%*3.98)*

The formula is based on the maximal binding of 2 mol Fe^{3+}/mol of transferrin and a molecular weight of 79,570gm/mol for transferrin (Morgan EH).

Biostatistical analysis:- The results were expressed as (mean±standard deviation). Pooled t-test was used for the comparison between the patients and control groups in the measured parameters. ANOVA test used to compare the results of male and female patients group.

Results:

The results of iron indices expressed as mean ± standard deviation are presented in Table (1). There is a significant increase ($p<0.05$) in all iron indices of hypertensive patients in comparing with healthy control group except TIBC, UIBC, and transferrin concentrations, which decrease in these patients in comparing with control group. All parameters are increased in hypertensive patients.

Table (1): Iron indices in patients and control groups

Iron indices		Patients Group	Control Group	p-Value
Hb	(g/dL)	13.44±2.11	12.65 ±1.13	0.007554
PCV	%	43.31± 6.32	40.94 ±3.39	0.007483
S.Ferritin	(pmol/L)	383.12 ± 421.60	164.34 ±115.49	3.228E-05
EIBS	(mmol/L)	22.21±25.68	10.46±7.35	3.640E-05
S.Iron	(umol/L)	21.67± 12.54	17.85 ±6.52	0.046
TIBC	(umol/l)	42.94±14.95	56.17 ± 9.96	2.273E-09
TS%		54.77±35.20	32.81±11.62	3.874E-09
Transferrin Conc. (g/L)		0.11±0.03	0.14 ±0.03	8.804E-12
UIBC	(umol/l)	23.97± 16.83	38.47 ±11.09	7.154E-08

Table (2) showed the comparison between iron indices in male and female patients. There is a significant increase in serum ferritin and EIBS in male group as compared with female group. While all other iron indices were differ insignificantly.

Table (2): Serum level of the measured parameters in male patients in comparison with female patients.

Iron indices		Male Group	Female Group	p-Value
Hb	(g/dL)	14.38 ±2.45	12.78 ± 1.86	0.094
PCV	%	45.69 ± 6.76	41.49 ± 5.59	0.246
Ferritin	(pmol/L)	455.71 ± 433.23	181.48 ± 124.02	3.763E-14
EIBS	(mmol)	28.15 ± 27.40	11.55 ± 7.89	3.763E-14

S.Iron	(umol/L)	18.67± 7.80	17.88 ± 6.79	0.458
TIBC	(umol/L)	39.67± 9.17	56.19 ± 10.76	0.407
TS	%	48.35 ± 20.40	32.91±11.11	0.397
Transferrin	(g/L)	0.10 ± 0.02	0.15 ± 0.03	0.578
UIBC	(umol/L)	21.69 ± 11.66	38.56 ± 11.34	0.873

Discussion:

High level of iron indices in patients group in comparing with control group (Table (1)) are in accordance with many other researches (Danesh J. Coronary Bozzini C., Girelli D). Although several studies found no association between iron stores and coronary artery disease (Sempos CT, Rauramaa R). Many epidemiologic studies have found a positive relationship between body iron stores and coronary artery disease (Kiechl S, Willeit Tuomainen TP,). However, lack of consistency in the epidemiologic studies is probably explained by the large variability in estimates of iron stores and iron intake and by the diversity of study outcomes (Corti MC, Gaziano). Large differences in incidence and mortality for coronary artery diseases between countries have been widely documented (Verschuren WM).

Plasma ferritin, transferrin saturation, total iron-binding capacity, or serum iron were used as objective markers for body iron stores. Of these biomarkers or indices, ferritin is considered the best single indicator of total body iron (Cook JD, Lipschitz). Hence, the EIBS, calculated from serum ferritin reflects iron store in addition to be the best indicator of total body iron. Iron catalyzes the formation of reactive oxygen species through the Fenton and Haber–Weiss reactions (Navab M, Fogelman. Free radicals cause lipid peroxidation, leading to the modification of LDL at the molecular level, facilitating its deposition and leading to the formation of atherosclerotic plaque (Horwitz LD). Generation of reactive oxygen species (Connolly ES Jr, Winfree CJ) has been proposed as a mechanism for reperfusion injury. Hydrogen peroxide (H_2O_2) and superoxide radical ($\cdot O^{2-}$) are generated during ischemia and reperfusion (Horwitz LD, Leff JA. Catalase). In the absence of iron, these species are weak oxidants that are inactivated by endogenous scavenger enzymes (31). However, iron, through the Fenton reaction, catalyzes production of hydroxyl radical ($\cdot OH$) from H_2O_2 and ($\cdot O^{2-}$). Because there are no effective endogenous defenses against this highly reactive molecule, $\cdot OH$ is highly toxic to tissue. Intracellular, not extracellular, iron availability is critical to toxicity from $\cdot OH$ (Lesnefsky EJ, Ye J.). Prevention of $\cdot OH$ formation by iron chelation prevents damage to cultured myocardial cells by H_2O_2 (Byler RM, Sherman). In the myocardial cell membrane, $\cdot OH$ can initiate lipid peroxidation or oxidation of protein sulfhydryl groups (Lesnefsky EJ, Dauber). Lipid peroxides are known to cause alterations in membrane permeability and enzyme activity in subcellular organelles. Lipid peroxidation causing lysozome membrane fragility and leakage of hydrolytic enzymes may ultimately lead to cell damage and death. The most acceptable explanation from the above discussions and references is that the increase in iron level causes oxidation and damage to artery wall leading to increase risk of placing cholesterol in walls and subsequently increase risk of coronary artery disease.

The results in Table (2) showed no significant differences between male and female groups in most iron indices except in ferritin and subsequently EIBS level.

This results may be explained by the fact that most of our female patients were in menopause state leading to lack of blood in menses. Therefore the iron level will be not affected by the menses although transferring level changed. A study by Lauffer (1991) (Lauffer RB.) showed a significant correlation between iron stores and cardiovascular mortality. In men, iron stores, assessed by serum ferritin concentration (Cook JD, Lipschitz) rise after adolescence. In women, iron stores remain low and only begin to rise after the age of 45 years (37). In young men, there is a parallel increase in risk of coronary heart disease and iron load (Sullivan JL). The maximal sex difference in serum ferritin level is reached at approximately 45 years of age and is about 300%. The maximal sex difference in heart disease is also reached at approximately 45 years and is also about 300% (Sullivan JL).

Conclusion:

It could be concluded that hypertensive patients have significantly high level of iron parameters than control group. There is no significant difference between male and female groups in most iron indices except in ferritin and subsequently EIBS levels. Further studies required for other biochemical parameters in larger patients sample size. Increased iron level causes oxidation and damage to artery wall leading to increase risk of placing cholesterol in walls and subsequently increase risk of coronary artery disease.

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دراسة حالة الحديد في مصول المرضى المصابين بارتفاع ضغط الدم الابتدائي

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الخلاصة:

يعد ارتفاع ضغط الدم من المشاكل الصحية الرئيسية عند البالغين ويساهم في احداث أمراض القلب والشرايين. ومقارنته بمستوياتهما عند الناس الأصحاء. يمثل الحديد في الجسم تناقض في أهميته من حيث انه يدخل في الكثير من العمليات الحيوية لكنه يمكن أن يكون مضرا. اجريت هذه الدراسة لمحاولة لتحديد وضع الحديد عند مرضى الضغط مستهدفين التوصل إلى تحديد وجود ارتفاع في مستوى الحديد كمؤشر للإصابة بأمراض القلب والشرايين عند مرضى الضغط.

شملت عينات الدراسة 88 شخصا مصابا بمرض ضغط الدم فقط وليس لديهم أمراض أخرى ومعدل أعمارهم بين (13±49سنة). تم قياس تراكيز الحديد والسعة الكلية للارتباط بالحديد (TIBC) والفيرتين بالطرق اللونية باستعمال العدد القياسية الجاهزة. حسبت سعة عدم الارتباط بالحديد(UIBC) والحديد المخزون الكلي بالجسم ونسبة تشبع الترانسفرين وتركيز الترانسفرين رياضيا باستخدام المعادلات الرياضية.

أظهرت النتائج ارتفاعا معنويا ($p<0.05$) في مؤشرات الحديد لدى المرضى مقارنة بمجموعة السيطرة عدا تراكيز الترانسفرين وUIBC وTIBC. لا يوجد اختلاف معنويا ($p>0.05$) في تراكيز مؤشرات الحديد عند المقارنة بين المرضى الذكور والإناث عدا الفيرتين و الحديد المخزون الكلي. *يستنتج* من هذه الدراسة أن مرضى ضغط الدم لديهم تركيز أكثر من الحديد في أجسامهم ولا توجد فروقات في ذلك بين الذكور والإناث عدا في الفيريتين. هذه النتائج تحتاج ان تدرس معها متغيرات كيموحياتية اخرى ولعينة اكبر من المرضى.

مفاتيح الكلمات: ارتفاع ضغط الدم، الحديد، الفريتين، TIBC