Study of the Correlation Between Serum C-reactive Protein and Bone Biochemical Markers in Type 2 Diabetic Individuals.

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Abstract:

Objective:- Diabetes Mellitus is a metabolic disorder and associated with number of disorders including bone, kidney ,liver, nerves, hart and blood vessels dysfunction. C- reactive protein (CRP) is a sensitive physiological biomarker of subclinical inflammation associated with hyperglycemia. The objective of the present study was to establish the relationship between CRP with blood sugar, Calcium (Ca^{+2}), Phosphorus (PO_4^{-2}) and Alkaline Phosphatase (ALP) in serum of Diabetes Mellitus type 2(DM2). **Methods:**- The study involved of 40 DM2 patients and 30 non-diabetic individuals as control. The colorimetric analysis has been applied for blood sugar, Ca^{+2}, PO_4^{-2}, ALP and CRP. Samples were collected from February 2013 to August 2013 of Tikrit Teaching Hospital.

Results and Conclusions:- There is a significant increase in(blood sugar, PO_4^{-2}, ALP and CRP) while (Ca^{+2}) level was observed significantly decrease in serum of DM2 patients compared with control. Correlation between CRP with blood sugar, Ca^{+2} and PO_4^{-2} was positive, while it was negative with ALP in DM2 compared with positive correlation observed between CRP with blood sugar and ALP but it was negative with Ca^{+2} and PO_4^{-2} in control group. Our conclusion is CRP has strong association with blood sugar, Ca^{+2}, PO_4^{-2} and ALP in DM2 and should looked for to have better control of blood sugar.

Key words:- CRP, Calcium, Phosphorus, Alkaline Phosphatase, Diabetes Mellitus.

Introduction:

Diabetes is a metabolic – disorder with inappropriate hyperglycemia either due to an absolute or relative deficiency of insulin secretion or reduction in the biologic effectiveness of insulin or both. It is also associated with disturbance concerned with protein, carbohydrate and lipid metabolism. The decreased uptake of glucose into muscle and adipose tissue leads to chronic extra cellular hyperglycemia which results in tissue damage and chronic vascular complications[(1)].

The international diabetic federation (IDF) estimated in 2011 that 366 million adults aged 20-79 years, of the world's 7 billion population have diabetes mellitus. This gives a comparative prevalence of 8.5%. Since more than 90% of the global cases of Diabetes mellitus are type 2, it is evident that the epidemic was mainly due to the escalation of the causes of Diabetes mellitus type 2 (DM2)[(2-4)].

Among several markers of inflammation, CRP is found to be significant in people with diabetes. CRP, a pentameric protein produced by the liver has emerged as the "golden marker for inflammation". It is a non-immunoglobin protein having five identical subunits. It is a member of pentraxin family protein. The C-reactive protein (CRP) derives from the fact that it reacts with capsule polysaccharide of *streptococcus pneumonia.* It is an acute phase response protein markedly increased in both inflammatory and infection diseases. It plays an important role in innate immunity. Experimental evidence demonstrated that CRP as a sensitive physiological marker of sub clinical systemic inflammation is associated with hyperglycemia, insulin resistance and overt DM2 [(1,5)].

Insulin, together with insulin-like growth factor; stimulates bone matrix synthesis. The stimulatory effect of insulin on bone matrix results from its action on the differentiating function of osteoblasts. Insulin is also necessary for normal bone mineralization. Osteoblasts control mineralization by regulating the passage of calcium (Ca^{+2}) and phosphate ion (PO_4^{-2}) across their cell membrane; these cells contain alkaline phosphatase (ALP) that is used to generate PO_4^{-2} from inorganic phosphates[(6-9)].

It has been reported that DM and connected with the disease metabolic disturbances lead to important alterations in bone metabolism. It has been reported that poorly controlled DM2 may be consequence of increased osteolysis and can lead to increased susceptibility to bone loss and development of bone changes as osteopenia. In the majority of recent studies it has been reported that bone turnover in patients with DM2 is decreased due to the decrease in the number of osteoclasts and delay in formation and mineralization of the osteoid[(10)] .The aim of this study was evaluation the association of CRP with biochemical markers blood sugar, Ca^{+2}, PO_4^{-2} and ALP in patients with DM2.

Materials and Methods:

This study included 40 patients with DM2 consisted of (22 women and 18 men) and were confirmed to be diabetic with bone pain, also 30 were age matched apparently healthy control subject on routine checkup, they were confirmed to be non diabetic subjects diagnosed with other systemic diseases were excluded . Samples were collected from diabetic patients admitted to Tikrit Teaching Hospital in Tikrit city from February 2013 to August 2013 with age ranges from (30-45) year with patients of Type2 D.M. Studies were preformed on peripheral venous blood samples withdrawn from the cubical vein in fasting condition. Blood samples 20-30 minutes after

collection were centrifuged for 10 minutes at 3000 rpm and the serum was separated to the plan tubes and stored at -20°C until assayed.

Fasting glucose, Calcium, Phosphorus and ALP activity was determined by the enzymatic colorimetric method (biolabo kit, France).CRP was determined turbidimetrically by using CRP (Spectrum kit, Germany).

Data were presented as Mean± Standard Deviation (SD) and analyzed using one way analysis of variance (ANOVA). Using SPSS version 18, P values 0.05 were considered significant. Correlation between analyzed variables were assessed with the use of Pearson's.

Results:

The mean ± SD of blood sugar in diabetic patients was (298±104 mg/dl) with high significant ($p\leq 0.01$) compared with control subjects (101.7±17.8mg/dl). Serum levels of Ca^{+2}, ALP and CRP in DM2 were (8.55±0.587 mg/dl), (14.62±4.32 K.A.U/dl), and (7.14±1.82mg/dl) respectively also were highly significant ($p\leq 0.01$) in comparison with the mean values of the control group were (9.24±0.516 mg/dl), (8.68±2.38 K.A.U/dl) and (5.02±1.12 mg/dl) respectively. The mean ± SD for serum PO_4^{-2} in DM2 was (4.045±0.395 mg/dl) showed a significant relation ($p\leq 0.05$) compared with controls (3.77±0.511 mg/dl). Biochemical characteristics of the study population are presented in table 1.

Table1:-The mean±SD for biochemical characteristics of the study population.

parameters	mean ± SD		P≤
	Diabetic individual	Control individual	
Blood sugar (mg/dl)	298±104	101.7±17.8	0.01
Calcium (mg/dl)	8.552±0.587	9.243±0.516	0.01
Phosphorus (mg/dl)	4.045±0.395	3.777±0.511	0.05
ALP (K.A.U/dl)	14.62±4.32	8.68±2.38	0.01
CRP (mg/dl)	7.14±1.82	5.02±1.12	0.01

The results of the Pearson's correlation analysis between the variables in control and DM2. There was a positive correlation between CRP with blood sugar and ALP (0.143 and 0.146) respectively (fig.1 and 2) while a negative correlation between CRP with Ca^{+2} and PO_4^{-2} (-0.266 and -0.322) respectively (fig.3 and 4) in control group, but a positive correlation between CRP with blood sugar, Ca^{+2} and PO_4^{-2} (0.978, 0.074 and 0.108) respectively (5, 6 and 7) while a negative correlation between CRP with ALP (-0.222) (fig. 8) in diabetic patients.

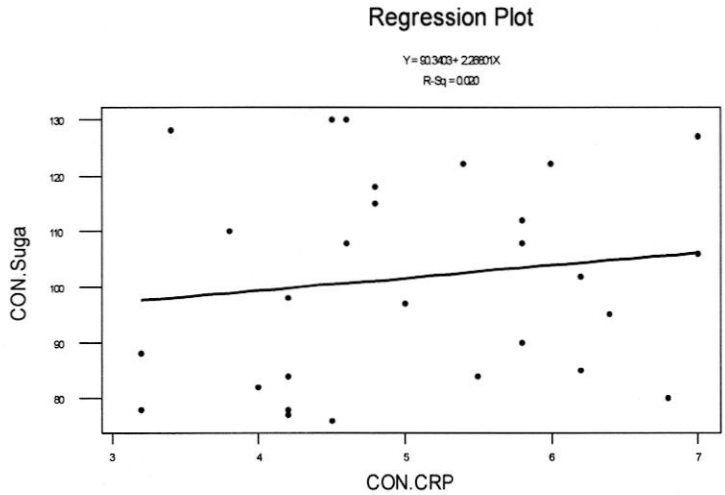

Fig (1):- Correlation between CRP with blood sugar in control group.

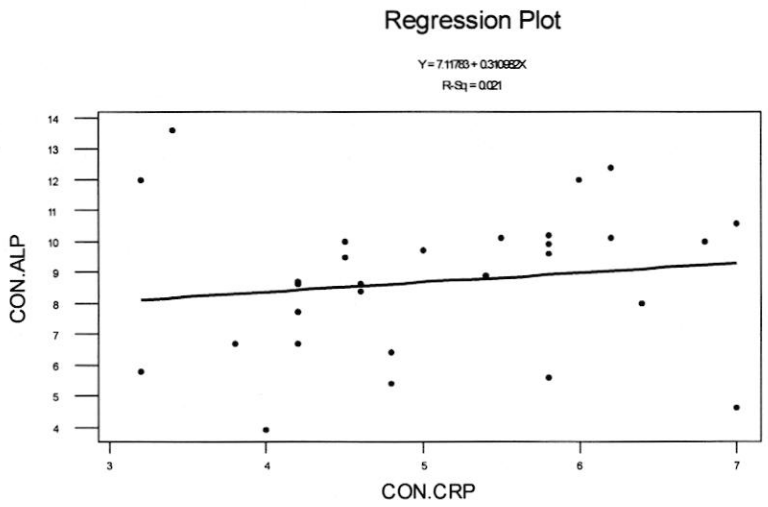

Fig (2):- Correlation between CRP with ALP in control group.

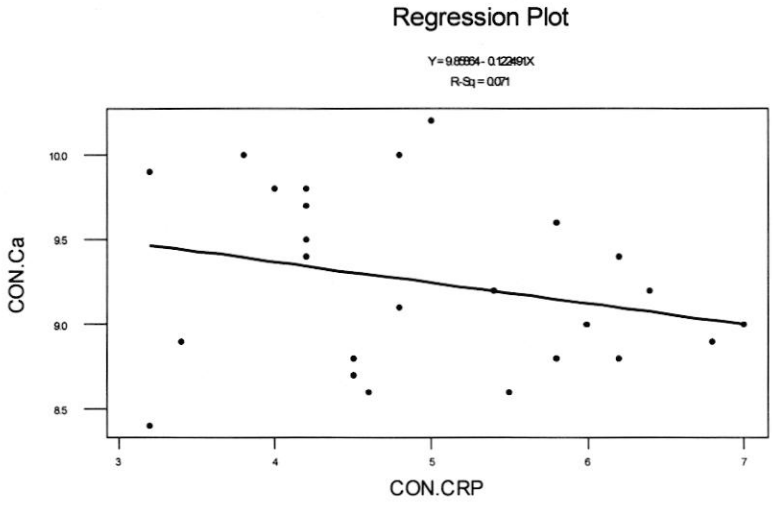

Fig (3):- Correlation between CRP with Calcium in control group.

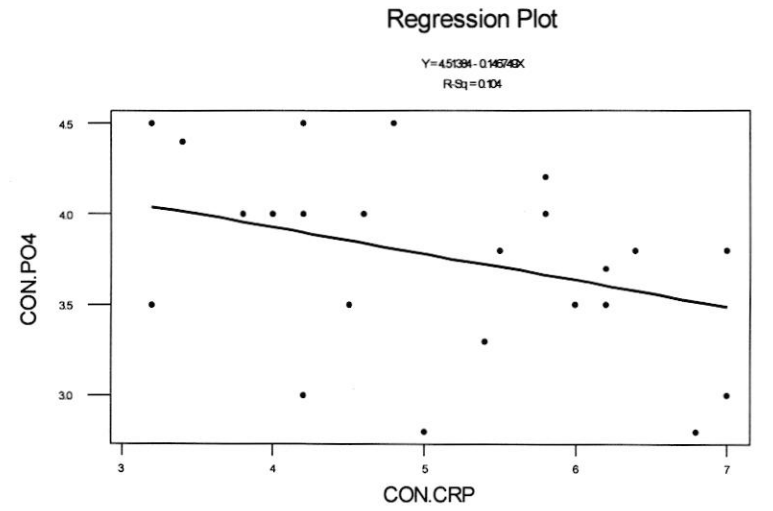

Fig (4):- Correlation between CRP with Phosphorus in control group.

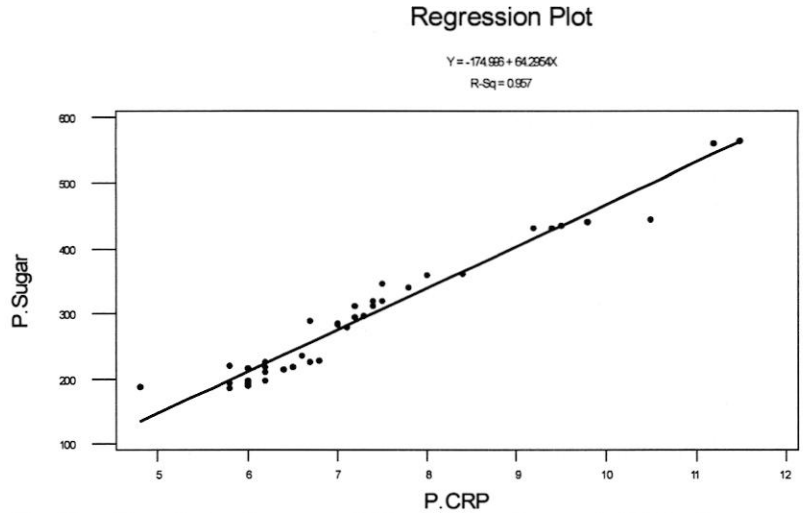

Fig (5):- Correlation between CRP with blood sugar in Diabetic group.

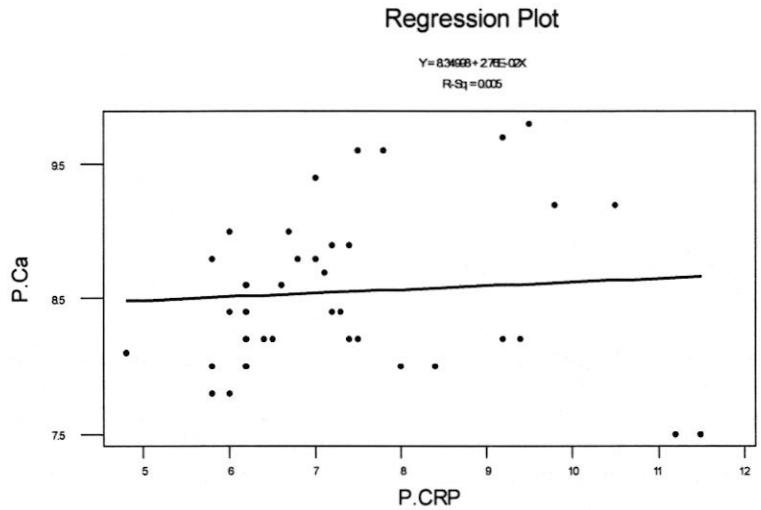

Fig (6):- Correlation between CRP with Calcium in Diabetic group.

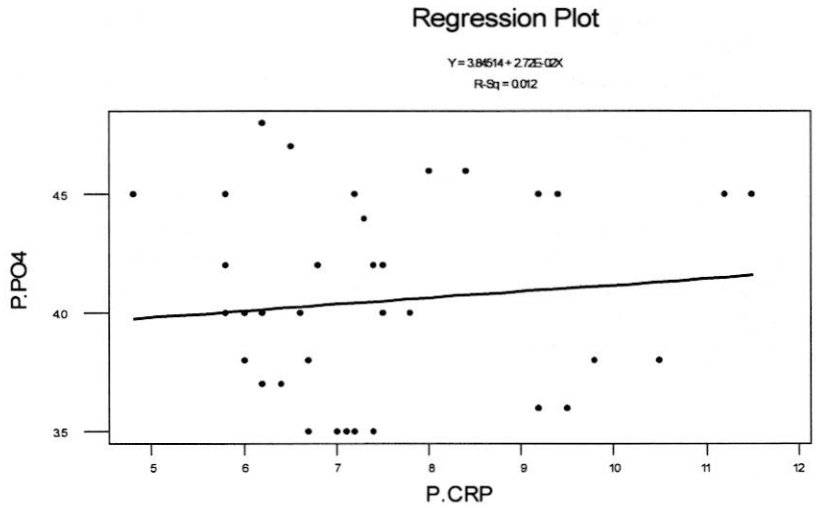

Fig (7):- Correlation between CRP with Phosphorus in Diabetic group.

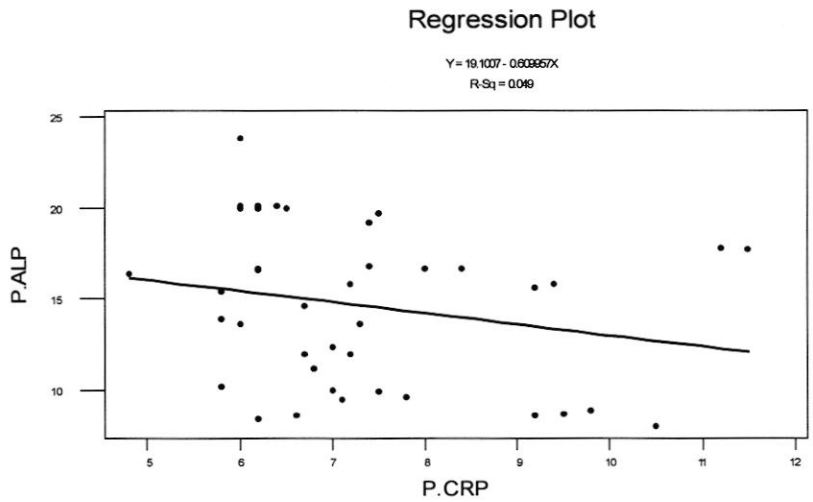

Fig (8):- Correlation between CRP with ALP in Diabetic group.

Discussion:

CRP an acute reactant produced by liver, is an extremely sensitive marker of systemic inflammation. It is perceived that chronic low-grade inflammation as evidenced by elevated high-sensitivity C-reactive protein (hs-CRP) might potentially be a cause underlying the etiology and manifestation of DM2[2,5]. The results in the study indicate a high significant increase in CRP, blood sugar and ALP in patients with control ($p \leq 0.01$). The findings are in agreement with previous findings [10-13].

It has been established that elevated cytosolic Ca^{+2} concentrations is the primary trigger for insulin release. However, reduced Ca concentrations in the lumen of acidic a compartment was also shown to inhibit exocytosis in the INS-1 β cell line. Indeed, insulin is released from pancreatic secretory β-cell, both under basal condition and in response to glucose secretion is defective in type 2 diabetes [12,14]. These finding are in agreement with some authors[10,12,14], others repotted opposite findings[10,15-17], while Hamed et al stated that levels of serum Ca between patients and control remained unaltered without significant effect of hyperglycemia accompanying diabetes[6]. Sun et al find significant positive correlations of serum glucose with calcium in women and men ($r=0.31$and 0.22 respectively; $p<0.001$ for both)[13]. Changes in Ca^{+2} is primary insulin target tissues may contribute to peripheral insulin resistance via impaired insulin signal transduction, leading to decrease glucose transporer-4 activity [18]. Also insulin influence the phosphorous execration, the higher the level of plasma glucose the higher the level of phosphorus [14] and it was shown that insulin has influence on diabetic bone metabolism. On the other hand amylin co- recreated with insulin by pancreatic β-cell. Its absence in diabetic is frequently associated with Osteopina. Amylin binds to calcium receptors, lowers plasma calcium concentration, inhibits osteoclast activity, and stimulates osteoblasts[9,19-20].

Bone- specific (ALP) is a key enzyme in osteolast for bone mineralization, most of the body's PO_4^{-2} is combined with Ca^{+2} in the bone, but about 15% exists in the blood and other soft tissues and body fluid. Biochemical markers of bone turnover released to circulation during metabolic activity of estrogenic cell-osteoblasts and osteolysis cell-osteoclasta which is effected in DM2 [10, 14].

It is beloved that inflammation has a crucial intermediary role in the pathogenesis of type2 Diabetic, thereby linking Diabetes Mellitus with a number of commonly co-existing condition through to originate via inflammatory mechanism. Recent research suggests that patients with elevated basal level of CRP are at an increased risk of Diabetes Mellitus, Hypertension and cardiovascular disease [2].

Conclusion:

C-reactive protein positively correlated with blood sugar, Ca^{+2} and PO_4^{-2} while it was negative with ALP level in serum of Diabetes type 2 compared with control group which showed positive correlation between CRP were negative with Ca^{+2} and PO_4^{-2} levels.

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دراسة الارتباط بين بروتين-C- المتفاعل مع المتغيرات الكيميائية الحيوية للعظام في الاشخاص المصابين بمرض السكري النوع الثاني

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الملخص:

الهدف: مرض السكري هو خلل ايضي يقترن مع عدة متغيرات في العظام ، ألكبد ، ألكلية الأعصاب القلب واعتلال الاوعية الدموية. يعتبر بروتين -C- المتفاعل (CRP) متغير حيوي حساس فسيولوجيا للالتهابات المرتبطة بارتفاع السكر. الهدف من الدراسة الحالية هوايجاد العلاقة بين CRP مع مستوى سكر الدم ، الكالسيوم (Ca^{+2})، الفسفور(PO_4^{-2}) و انزيم الفوسفاتيز القاعدي (ALP) في مصل المرضى بمرض السكري من النوع الثاني (DM2).

ألطريقة تضمن في الدراسة 40 مريضا مصابا بمرض السكري من النوع الثاني بالإضافة الى 30 شخص اصحاء بوصفهم مجموعة سيطرة وقد طبقت الطريقة اللونية لتحليل سكر الدم ، Ca^{+2} , PO_4^{-2} ,ALP و CRP.جمعت العينات من مرضى مستشفى تكريت التعليمي للفترة من شباط الى اب 2013 .

النتائج و ألاستنتاجات :وجد ارتفاع معنوية في مستويات سكر الدم ،PO_4^{-2} و ALP و CRP بينما لوحظ انخفاض معنوي في مستوى Ca^{+2} في مصل مرضى السكري النوع الثاني مقارنة مع مجموعة السيطرة. الارتباط بين CRP مع سكر الدم ،Ca^{+2} و PO_4^{-2} كان موجبا بينما كان سالبا مع ALP في مجموعة المرضى مقارنة مع مجموعة السيطرة حيث كانت العلاقة الارتباطية موجبة بين CRP مع مستوى سكر الدم و ALP في حين انها كانت سالبة مع كل من Ca^{+2} وPO_4^{-2} . يستنتج ان CRP يرتبط بقوة مع مستوى سكر الدم ،Ca^{+2}، PO_4^{-2} و ALP في مرضى السكري من النوع الثاني لذا يجب تقييم هذه النسب في الدم وتنظيمها لأغراض السيطرة على مستوى سكر الدم.

الكلمات الدالة:بروتين-C -المتفاعل،كالسيوم، فسفور ،انزيم الفوسفاتيز القاعدي ،مرض السكري.