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ORIGINAL ARTICLE

Relationship Between Protein Kinase C and Diabetic Nephropathy Patients in Al Anbar City

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ABSTRACT

Background : Diabetic syndrome is characterized by increase blood sugar and resistance to insulin. It is linked to macrovascular and microvascular complications such as nephropathy, neuropathy, and retinopathy **.** the aim of this study to investigate the relationship protein kinase C and diabetic nephropathy.

Material and Methods: The study was include 84 samples , 36 of them with type 2 diabetes, 23 of them with diabetic nephropathy, and 25 healthy controls population , all of whom were over the age of 40. ELISA was used to calculate the protein kinase C level, fasting serum sugar , Glycated Hemoglobin ,Urea, Creatinine, Total protein, Albumin, and globulin were all found. Body mass index was also calculated.

Result : the study result shown approximately same level between healthy and patients groups and this also associated with increase in fasting serum sugar , Glycated Hemoglobin , Urea , Creatinine and body mass index in patients group compared to Healthy controls group.

Conclusion: serum protein kinase C shown non-significant difference between healthy and control patients.

Keywords: protein kinase C , diabetes , chronic kidney diseases anthropometric measurement .

INTRODUCTION

Diabetic nephropathy is one of the cause that lead to end-stage renal disease (ESRD) worldwide(1), and it is risk factor for cardiovascular mortality in diabetic patients (2) . patients who have type I diabetes , 25–40% of them will develop diabetic kidney disease (nephropathy) (3) .In patients how have type II diabetes 5% of them will have diabetic nephropathy at time of diagnosis and other 30–40% will develop diabetic nephropathy with a high risk of progression to ESRD(4). With the continuous increasing in the population how have type II diabetes , it is necessary to know the pathophysiology for this disease in order to design specific treatment for prevention and reversal of diabetic kidney disease.

Increase blood sugar shown to be the risk factor responsible for the development and progression of micro vascular complications of diabetes (5). "The Diabetes Control and Complications Trial in type I diabetes"(6) and the "United Kingdom Prospective Diabetes Study in type II diabetes"(7) shown that intensive control of blood sugar successfully delayed the onset and progression of diabetic micro vascular complications such as nephropathy, neuropathy , retinopathy. These data suggested that high blood glucose induce metabolic effect that cause theses vascular complications . Many biochemical pathways have been suggested to demonstrate the adverse effects of hyperglycemia. Activation of diacylglycerol (DAG)- protein kinase C (PKC) pathway (8) , enhance polyol pathway,(9) increased oxidative stress (10) and overproduction of advanced glycation end products (11), all these suggested as potential cellular mechanisms by which hyperglycemia induces vascular complications in diabetic.

Protein kinase is family of serine – threonine (EC 2.7.11.1) that consist of twelve isoforms ,that are classified according to whether they contain domains that bind Ca^{2+} (calcium) or DAG (diacylglycerol), both of it regulate the kinase activity positively . Conventional PKC are (α, β1, β2, and ϒ) binds to both Ca^{2+} and DAG, while novel PKC (☐, e , Z , y, m) binds to DAG, but not to Ca^{2+}, and atypical PKC (z , l) that not bind to any of these . The activation of novel and conventional PCK isoforms require phosphorylation of these isoforms and cofactors like Ca^{2+} and DAG. When its phosphorylated properly, increases Ca^{2+} or DAG chronically or rapidly will induce it`s translocation to the membranous compartment of the cell to perform the biological action . cytokines usually induce rapid and short term increase in Ca^{+} and DAG levels by activation of phospholipase C . Chronic activation of PKCs need sustained elevations of DAG, which include the activation of phospholipase D/C or the de novo synthesis of DAG (12).

Activation of conserved hepatic aPKC in obesity and type 2 diabetes cause problem, as hyperinsulinemia activates hepatic aPKC and aPKC-dependent processes inordinately by: (1) activation of sterol receptor element binding protein-1c (SREBP-1c), which transactivates many of lipogenic genes, that include fatty acid synthase (FAS) and acetyl-CoA carboxylase (ACC) , and also by (2) activation inhibitor of κ-B kinase-ß (IKKß), that phosphorylates IκBß, the inhibitor of nuclear factor κB (NFκB), therefor releasing NFκB for nuclear uptake and transactivation of proinflammatory cytokine genes, including TNF-α and IL-1ß (13)

METHOD

This study included 84 people that divided into three groups : 25 healthy people control group (16 males and 9 females), 23 patients with renal failure (19 males and 4 females), and 36 patients with diabetes type 2 (17 males and 19 females). All patients and healthy control samples were chosen above the age of 40.

Statistical Analysis: The data was statistically analyzed using SPSS version 24 and GraphPad prism version 7. P value less than 0.05 chosen as significant difference . Descriptive statistics were calculated for each parameter, including mean and standard deviation (SD). The Student's t-test was used to compare T2DM and (chronic kidney disease) CKD. Using Pearson's correlation (r=1 to 1), the PKC characteristics of both T2DM and CKD was investigated. Receiver's operating characteristic(ROC) curve was developed to investigate the distinct ability of PKC levels in healthy individuals and patients with T2DM and CKD.

RESULT

The table one show the mean and standard deviation (SD) , and also show the p value were less than 0.05 have a significant difference .

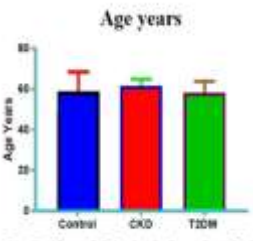


Fig. (1): Mean± S.D for Age in Control and Patients

Table 1: Clinical and Anthropometric characteristic of healthy control, CKD and T2DM patients

Parameter	Healthy controls		CKD Patients		T2DM Patients		p-value
	Mean	SD	Mean	SD	Mean	SD	
Age years	50.9	5.8	61.48	3.666	58.22	6.678	0.6782
BMI kg/m^2	25.6	2.205	26.8	4.256	29.33	5.006	0.0123
FSG mg/dL	115.7	10.58	211.3	40.58	202.8	57.24	<0.0001
HbA1c %	5.311	0.6231	6.358	1.283	9.244	2.433	<0.0001
Urea mg/dL	37.93	5.368	158.2	43.43	43.01	10.9	<0.0001
Creatinine mg/dL	0.536	0.1467	3.704	1.278	0.6636	0.171	<0.0001
T. proteins g/dL	8.728	0.6548	7.608	0.7192	8.626	1.121	<0.0001
Albumin g/dL	4.196	0.443	3.125	0.4264	3.967	0.5523	<0.0001
Globulins g/dL	4.533	0.7581	4.484	0.6863	4.659	1.026	0.7236
ALB/GLB	0.9581	0.2244	0.7219	0.1913	0.8967	0.2602	0.0016
Na Ions mmol/L	155.1	10.6	156.3	16.19	139.6	15.01	0.3472
K Ions mmol/L	4	0.6212	4.157	1.077	3.772	1.084	0.3173
Ca Ions mg/dL	9.181	1.562	8.308	1.754	8.917	1.711	0.1893
Uric Acid mg/dL	6.564	1.207	7.491	1.567	6.731	1.172	0.0304
Cr Cl mL/min	177	52.83	26.88	12.72	136.9	60.44	<0.0001
PKC	11.98	2.516	11.38	3.509	12.17	5.296	0.6395
IL-20 ng/mL	1.638	0.6743	0.826	0.2702	1.683	0.6103	<0.0001

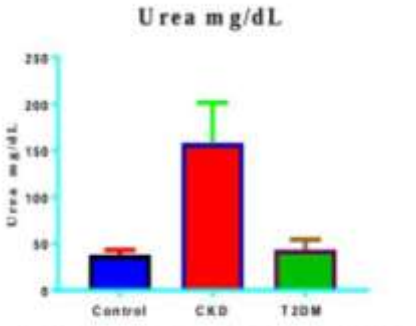


Fig. (5): Mean+ S.D for Urea in Control and Patients

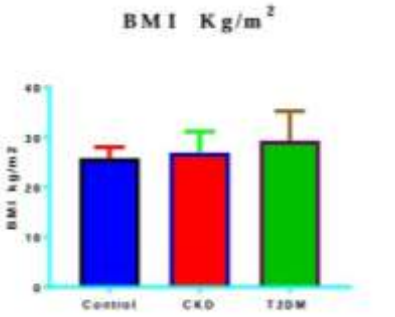


Fig. (2): Mean+ S.D for BMI in Control and Patients

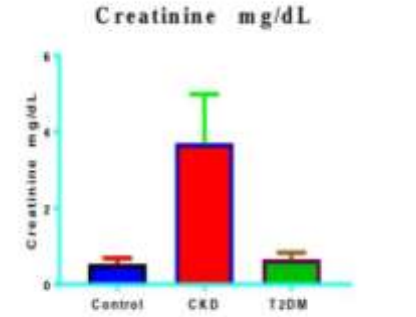


Fig. (6): Mean+ S.D for Creatinine in Control and Patients

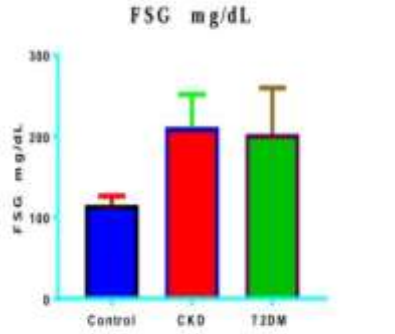


Fig. (3): Mean+ S.D for FSG in Control and Patients

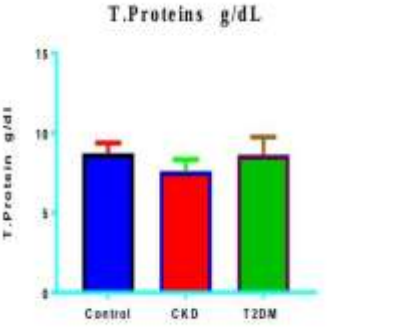


Fig. (7): Mean+ S.D for T.Protein in Control and Patients

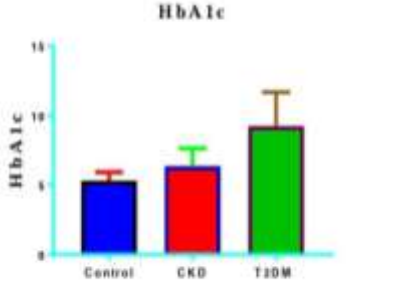


Fig. (4): Mean+ S.D for HbA1c in Control and Patients

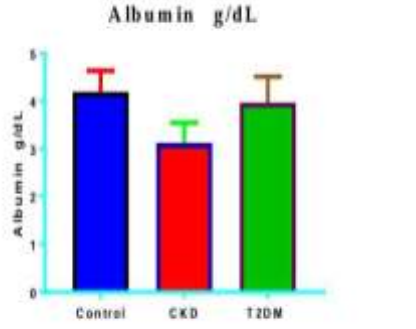


Fig. (8): Mean+ S.D for Albumin in Control and Patients

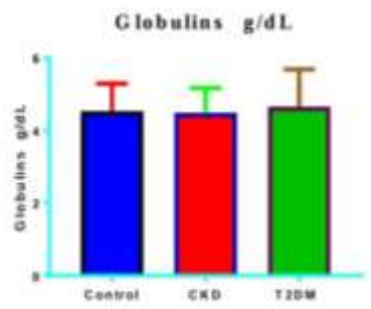


Fig. (9): Mean+ S.D for Globulins in Control and Patients

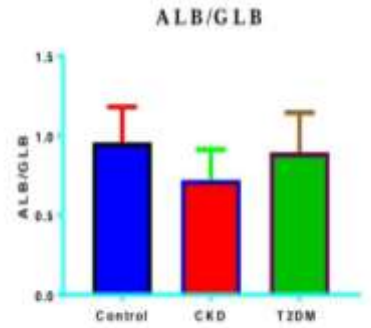


Fig. (10): Mean+ S.D for ALB/GLB in Control and Patients

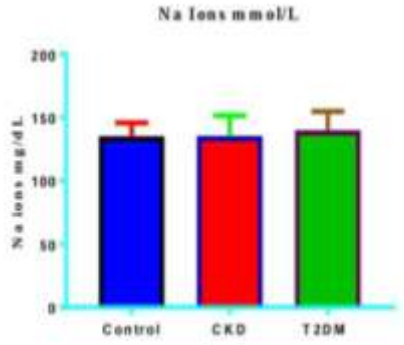


Fig. (11): Mean+ S.D for Na Ions in Control and Patients

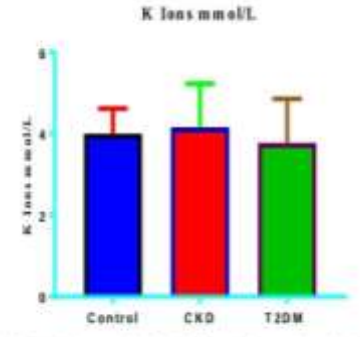


Fig. (12): Mean+ S.D for K Ions in Control and Patients

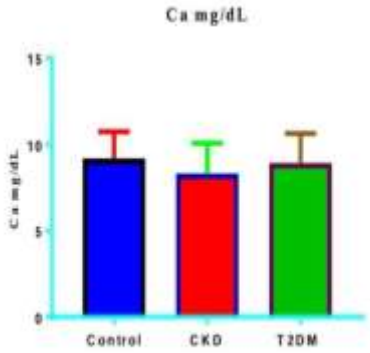


Fig. (13): Mean+ S.D for Ca in Control and Patients

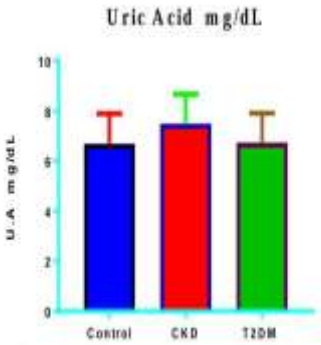


Fig. (14): Mean+ S.D for Uric Acid in Control and Patients

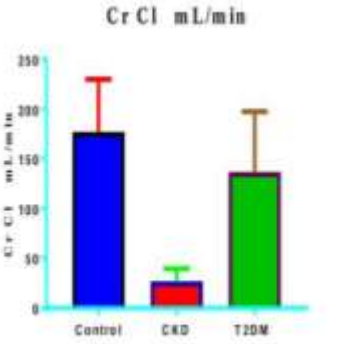


Fig. (15): Mean+ S.D for Cr Cl in Control and Patients

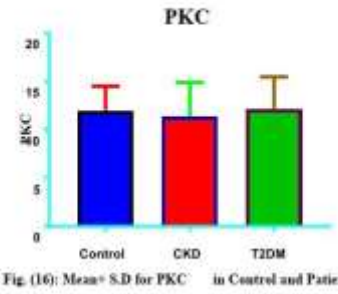


Fig. (16): Mean+ S.D for PKC in Control and Patients

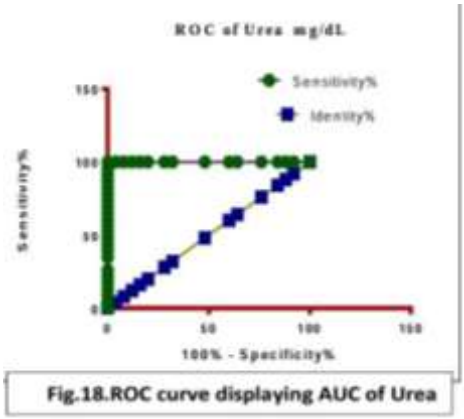


Fig.18.ROC curve displaying AUC of Urea

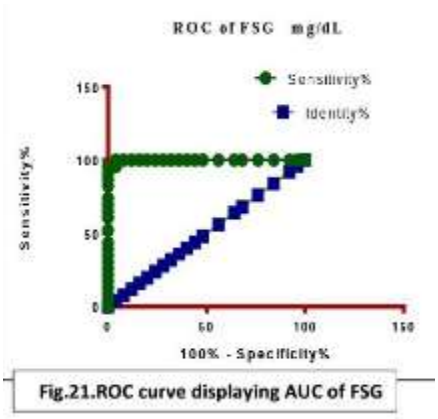
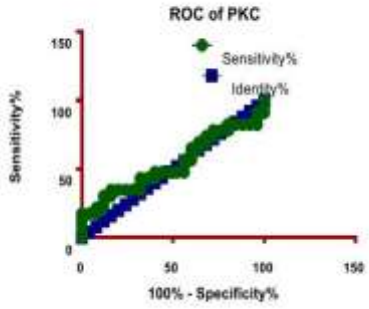


Fig.21.ROC curve displaying AUC of FSG



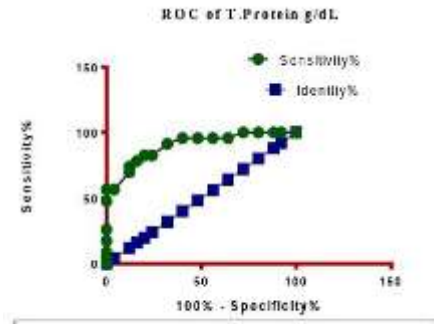


Fig.22.ROC curve displaying AUC of T.Protein

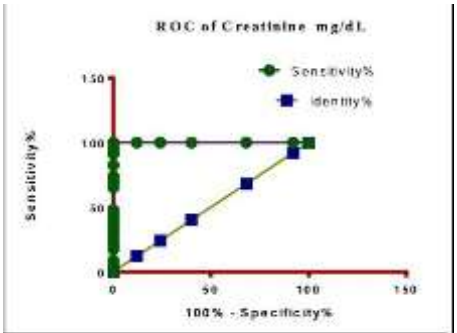


Fig.19.ROC curve displaying AUC of Creatinine

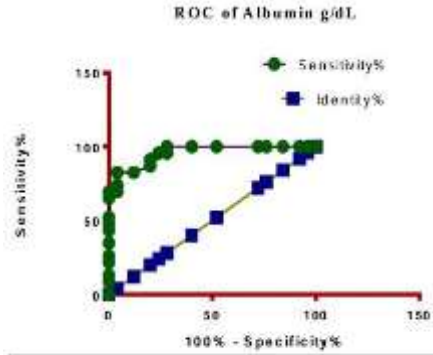


Fig.23.ROC curve displaying AUC of Albumin

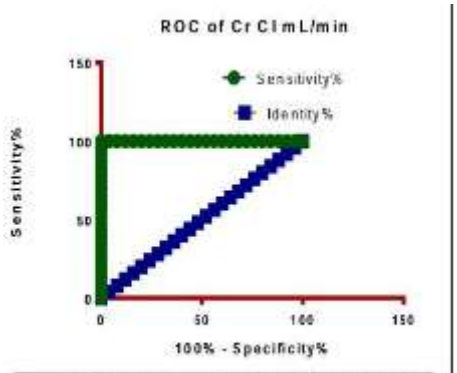


Fig.20.ROC curve displaying AUC of Cr Cl

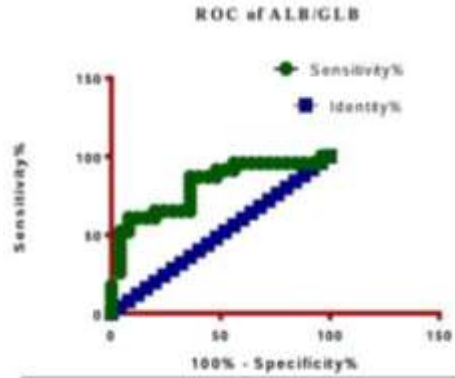


Fig.24.ROC curve displaying AUC of ALB/GLB

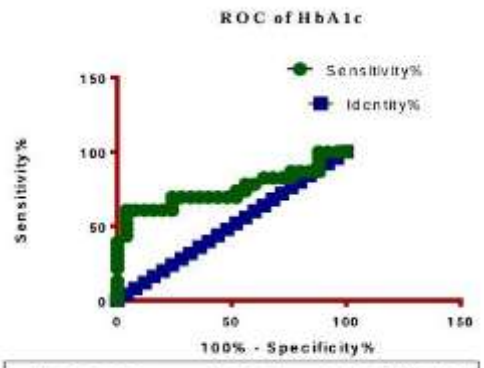


Fig.25.ROC curve displaying AUC of HbA1C

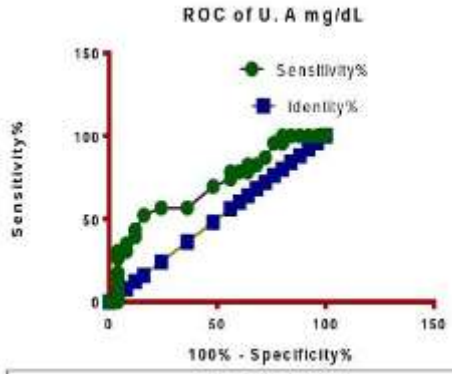


Fig.26.ROC curve displaying AUC of Uric Acid

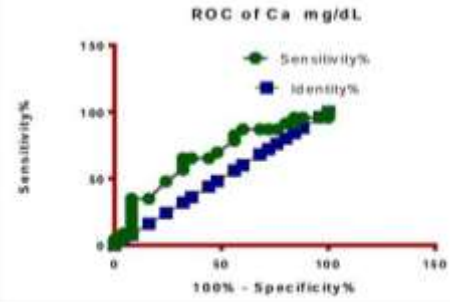


Fig.27.ROC curve displaying AUC of Calcium ion

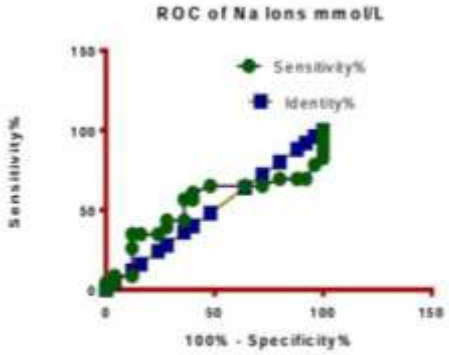


Fig.28.ROC curve displaying AUC of Sodium ion

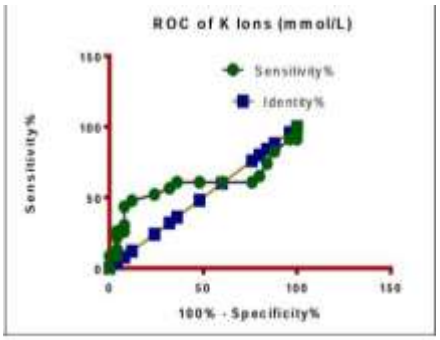


Fig.29.ROC curve displaying AUC of Potassium ion

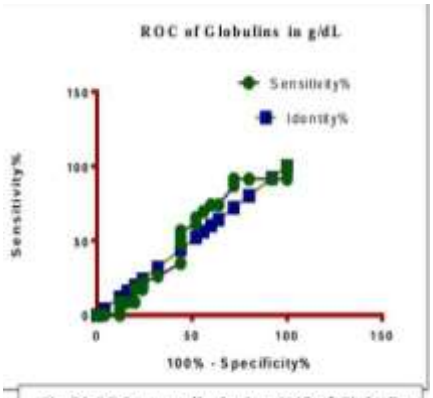


Fig.30.ROC curve displaying AUC of Globulin

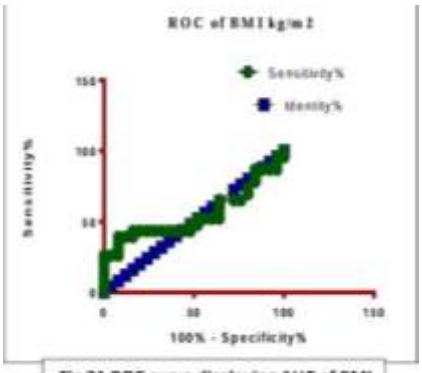


Fig.31.ROC curve displaying AUC of BMI

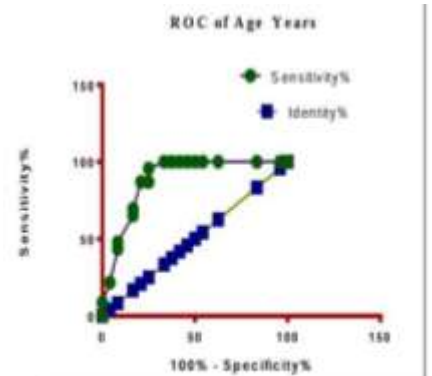


Fig.32.ROC curve displaying AUC of Age

Table 2: the correlation of protein kinase C with all parameters in this study

Correlation of PKC with all Studied Parameters		
Parameter	r (PKC pg/mL)	p-value
PKC	1.000	0.000
FSG mg/dL	-0.075	0.500
HbA1C	0.003	0.978
Urea	-0.139	0.208
Creatinine	0.005	0.964
Na+	-0.203	0.063
T.Protein	0.085	0.440
S.Ca	0.125	0.257
Albumin	-0.039	0.727
ALB/GLB	-0.118	0.288
globulins	0.129	0.243
K+	-0.074	0.504
BMI	-0.162	0.142
Cr. Cl.	-0.109	0.322
Uric acid	-0.026	0.813

DISCUSSION

Protein kinase Cs (PKCs) are particularly important mediators of immune intracellular signaling (14). this study shows that (PKCs) have non -significant difference between healthy and diabetic patients . this result was opposite to the result of some studies that shown activation in this pathway (5,8) , PKCs are important mediators of immune intracellular signaling (15), this result may be explained as diabetic mellitus impairs cellular functions such as phagocytosis, chemotaxis , and pathogen killing by macrophages, monocytes, and neutrophils. When diabetic monocytes, macrophages, and neutrophils were compared to control cells, most studies found a decrease in normal cellular function as well as changes in enzyme activity and cytokine secretion (16). the protein kinase dependent pathway play important role in synthesis of cytokines (17), low level of some cytokines shown in diabetic patients (18). There is an association between low capacity production of pro inflammatory cytokine and high level of blood glucose , high HBA1C , and dyslipidemia(19), that consist with this study finding as patient with type 2 DM have hyperglycemia, high HBA1c . This study also found that patients with T2DM have higher levels of serum urea and creatinine . as many research showed that urea and creatinine are useful predictor for renal failure in diabetic patient (20).this study found a decrease in total protein and albumin in diabetic and CKD, low protein diabetic patient indicate renal injury, this may due to change in basement membrane of renal glomeruli that will lead to leakage of albumin and some other proteins (21) The globulin show no significant difference and this result may be due to compensatory increase in globulin due to decrease albumin in renal injury (22). The uric acid have been shown significant difference ,with higher level among patient with chronic kidney disease, this also was the result of some research (23). The endothelial dysfunction was associated with reduced eGFR that leads to the retention of substances like uric acid (24). The sodium (Na^{+2}) , potassium (K^{+2}) and calcium (Ca^{+2}) show non-significant difference.

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