

# A Relationship of Periostin with some Bio- parameters in Type2 Diabetic Patients without and with COVID19



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## ABSTRACT

**Background:** Type 2 diabetes mellitus (T2DM) is a prolonged disease characterized by hyperglycemia resultant from weakening in insulin secretion, insulin action or both. T2DM and its associated metabolic disorders have been stated as the main cause in coronavirus disease 2019 (COVID-19). Current study was designed to investigate the association of periostin with some parameters in T2DM patients without and with COVID19.

**Materials and methods:** This study involved of 56 patients, 28 with T2DM without any complications, the other 28 have T2DM with COVID19 attending Al-Fallujah teaching hospital, and 28 healthy persons matching in age, gender and ethnic background as control group. Serum level of periostin was estimated by ELISA technique while urea and creatinine were assessed by colorimetric enzymatic methods. Total protein and albumin were determined by biuret method.

**Results:** Serum level of periostin was elevated in the T2DM patients without and with COVID19 than in healthy controls (HCs). The levels of urea and creatinine were importantly higher in T2DM patients without and with COVID 19 than in HCs ( $P < 0.0001$ ), while total protein and albumin were lower in T2DM patients without and with COVID 19 than in HCs ( $P < 0.0001$ ). Periostin had significant positive association with fasting serum glucose (FSG), HbA1c, urea creatinine, BMI, and ALB/GLB ( $P < 0.01$ ), while negative association between total protein and albumin ( $P < 0.01$ ) was observed. Studied biomarkers offered the following descending order of area under the receiver operating characteristic (AUROC) curve FSG and HbA1c (1), periostin (0.9764) , urea (0.6569), albumin and ALB/GLB (0.641), total protein (0.6147), creatinine (0.611) and globulins (0.611).

**Conclusion:** Serum level of periostin may be used as a novel biomarker in diagnosis T2DM without and with COVID19.

## Introduction

Type 2 diabetes is impairment in the way that the body regulates and uses sugar (glucose) as a fuel. This long-term (chronic) condition results in too much sugar circulating in the bloodstream. Eventually, high blood sugar levels can lead to disorders of the circulatory, nervous, and immune systems, since of different risks pretended by the coronavirus disease 2019 (COVID-19). Before the COVID-19 disease, common practice was to see T2DM patients in the hospitals each 5 months to asses biomarkers related to hyperglycemia and self-management advice to provide some new drugs. The prevention and control the type-2 diabetes by changing lifestyle and dietary, if diet and exercise alone fail to lower blood glucose type-2 diabetics require insulin or oral hypoglycemic agents (medication that helps lower blood sugar) [1].

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At the present time, the benefits of the visit must be re-evaluated in a balanced manner with the possibility of infection acquiring severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) when visiting medical clinics. These risks were discovered by evaluating that diabetic patients are the most vulnerable to infection with COVID19 and complications which may lead to death [2].

Periostin part of family of fascilin, expressed in many normal fetal and adult tissues, was firstly recognized in osteoblasts and initially cloned from an osteoblast cell line of mouse. Periostin is an extremely preserved extracellular matrix protein and contributes by active method in the tissue injury pathophysiology, tumor development, fibrosis

inflammatory diseases, and atherosclerosis [3]. Periostin is a protein expressed and secreted in fibroblasts cardiac muscles, contributed in dysfunction of cardiac muscles [4]. Overexpression and the change in the level of periostin was established in many diseases such as lung disease, cardiovascular diseases, and cancer [3, 5]. It may work as a remodeling protein in inflammatory responses by act as scaffold, insulin resistance, and obesity that could give a new molecular clue for the obesity and T2DM pathogenesis [6].

Medical data in 2018 for 287 patients with pancreatic cancer [7] showed that the concentration of periostin is increased in these patients as compared to HCs, and the early-stage patients have lower periostin concentrations than the end stages patients. Another study in 2017 with eighty pancreatic cancer patients exhibited that those with great expression of periostin had approximately larger tumor volumes by two-times or more and proposed the irregular upregulation triggers by periostin of cells of pancreatic cancer [8].

One of the previous researches detected a relationship between metabolic diseases and periostin and suggested that obese T2DM patients had higher serum levels of periostin, and it is related with insulin resistance serum TG and chronic inflammation [6]. Previous study was revealed a strong relationship between complications of diabetic vascular and periostin [9]. Moreover, periostin was increased in the membranes of neovascular and vitreous for T2DM patients [10], demonstrating that periostin could has a vital role in the T2DM pathogenesis.

Periostin has key role in many respiratory syndromes. Recent study shows that periostin may be used as indicator for progression of many diseases with fibrosis of pulmonary [11]. Periostin is a protein of matricellular. Proteins of matricellular are extracellular matrix (ECM) proteins with nonstructural properties, which greatly expressed at sites of inflammation or injury. They relate with many ECM proteins to bind to cytokines and growth factors to modulate their activities and remodelling other tissues [12]. Periostin expression, the human periostin encoding gene, can be prompted by the interleukin (IL)-4, IL-13, and IL-17 [13]. Periostin relates with many molecules work together in cascades of signal to control the expression of different genes like those encoding chemokines, transforming growth factor- $\beta$  (TGF- $\beta$ ), and collagen [14].

The main goal of this paper is to evaluate periostin serum levels in T2DM patients without and with COVID19, and to determine the correlation of periostin with some biomarkers in type 2 diabetic patients without and with COVID19.

### Materials and methods

This study included fifty six patients, twenty eight of them with T2DM without any complications (group B), the remaining twenty-eight had T2DM with COVID19 (Group C) which were diagnosed with quantitative RT-PCR and

computerized tomography (CT) scan or chest X-ray at the 7-12 day from symptoms on set. The diagnosis of patients with COVID19 was also done by assessing the serum levels of ferritin and D-dimer, mean duration of T2DM was  $10.45 \pm 3.89$  years, 65% of them was had Glucophage, 21% taking Amaryl and others taking sitagliptin, janumat and novonorm, twenty-eight person were recorded in the study as healthy controls (group A), matching with patients in age, sex and ethnic background, the age range for all subjects were within 35→65 years, randomly chosen from Al-Anbar province those attending in Al-Fallujah teaching hospital between November 2021 to January 2022. The serum was collected before the treatments, fasting serum glucose (FSG), urea, and creatinine were measured by enzymatic methods through using available commercial kits (Linear, Spain). Total protein and albumin were determined by biuret method, while the concentration of periostin was determined by ELISA kit (BT LAB Inc, China)

### Statistical analysis

Statistical analysis of data was completed through GraphPad Prism program version 8. The results are listed as mean  $\pm$  standard deviation (SD). The significance of statistical analysis of the variances among the cases were tested with one way ANOVA analysis. Pearson correlation coefficient was used to determine the association of Periostin with other studied parameters, while the accuracy of the examination was stated by area under curve (AUC). P-value equal or less than 0.05 was considered statistically significant.

### Results

From **Table No.1**, the mean age (years) was  $50.04 \pm 8.988$ ,  $52 \pm 8.628$  and  $54.31 \pm 6.898$  in group A, B and C, respectively ( $P=0.1505$ ). Serum level of urea and creatinine (mg/dL) were importantly lower in group A than in B and C groups ( $p < 0.0001$ ), patient groups had lower serum levels of total protein and albumin (g/dL) as compared to A group, with  $P < 0.05$ , while ALB/GLB has low levels in group A than in B and C groups ( $P = 0.0074$ ), but serum levels of periostin were lower in group A compared with B and C groups, ( $p \leq 0.0001$ ).

**Table 1: Data of Studied Parameters in T2DM Groups and HCs.**

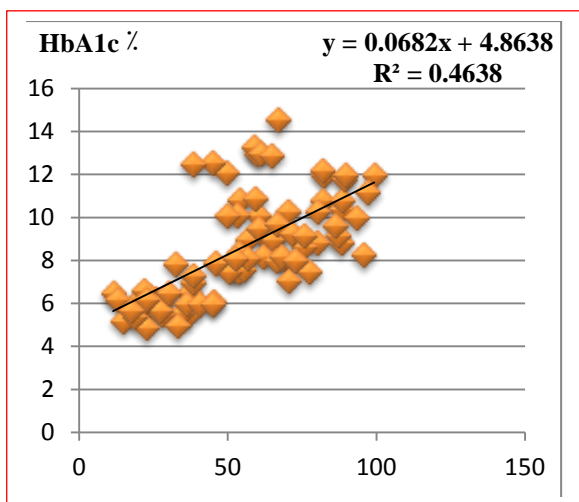
Parameters	Group (A)	Group (B)	Group (C)	p-value
	Mean $\pm$ SD			
The Age Years	50.04 $\pm$ 8.988	52 $\pm$ 8.628	54.31 $\pm$ 6.898	0.1505
FSG mg/dL	92.35 $\pm$ 9.277	181.9 $\pm$ 48.83	216.2 $\pm$ 59.94	<0.0001
HbA1c %	5.905 $\pm$ 0.4395	9.355 $\pm$ 2.186	10.18 $\pm$ 1.503	<0.0001
D-Dimer ng/mL	-----	-----	2257 $\pm$ 487.3	-----
Ferritin ng/mL	-----	-----	1279 $\pm$ 274.8	-----
Urea mg/dL	28.15 $\pm$ 6.887	33.65 $\pm$ 10.54	53.38 $\pm$ 13.74	<0.0001

Creatinine mg/dL	0.7918±0.2012	0.8568±0.2301	1.114±0.2939	<0.0001
T. proteins g/dL	8.456±0.7815	8.224±0.6714	7.737±1.264	0.0159
Albumin g/dL	5.086±0.5564	4.869±0.355	3.985±0.3889	<0.0001
Globulins g/dL	3.368±0.8889	3.355±0.7032	3.863±0.9065	0.0393
ALB/GLB	1.249±0.0324	1.26±0.0216	1.399±0.3358	0.0074
Periostin ng/mL	26.03±9.241	56.3±11.78	76.87±14.04	<0.0001

Significant strong positive correlation was sensed for periostin with FSG, HbA1c ( $r = 0.679$ ,  $P < 0.01$ ), ( $r = 0.681$ ,  $P < 0.01$ ), (Figures 1 and 2), moderate positive relationship of periostin with ALB/GLO, Urea, BMI ( $r = 0.5278$ ,  $P < 0.01$ ), ( $r = 0.509$ ,  $P < 0.01$ ), ( $r = -0.499$ ,  $P < .01$ ), respectively and weak positive correlation was noticed for periostin with creatinine, globulins ( $r=0.359$ ,  $P<0.01$ ), ( $r=0.113$ ,  $P=0.306$ ), respectively, also inverse association between albumin and total protein with periostin ( $r=-0.542$ ,  $P<0.01$ ), ( $r=-0.301$ ,  $P<0.01$ ), respectively was noticed as shown in table 2.

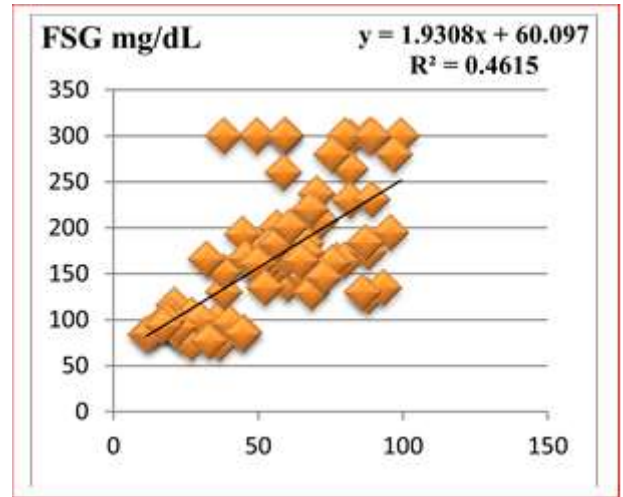
**Table 2: Association of Periostin Level with Studied Parameters**

	Periostin ng/mL	p-value
Periostin ng/mL	1.000	0.00
FSG mg/dL	0.679	<0.01
Hb A1c %	0.681	< 0.01
BMI kg/m <sup>2</sup>	0.499	< 0.01
Urea mg /dL	0.509	< 0.01
Creatinine mg/dL	0.359	< 0.01
Total protein g /dL	-0.301	< 0.01
Albumin g /dL	-0.542	< 0.01
Globulins g /dL	0.113	0.306
ALB/GLO	0.5278	<0.01



Periostin ng/mL

**Figure 1: Correlation of Periostin Level with FSG**  
Periostin ng/mL



**Figure 2: Correlation of Periostin Level with HbA1c**

ROC curve investigation exhibited that the best parameters had the ability to discriminate controls from T2DM patients were FSG and HbA1c [AUC=1;  $P < 0.0001$ ; 95% confidence interval (CI): 1 to 1 and SE: 0] as presented in Table 3, also periostin was establish to be the best parameter to predicate and diagnose T2DM [AUC=0.9764;  $P < 0.0001$ ; 95% CI: 0.9459 to 1.007 and SE:0.01554] as exposed in Table 3 and urea [AUC=0.6569;  $P < 0.0001$ ; 95% CI: 0.5133 to 0.8004 and SE: 0.07324] as displayed in Table 3. Similarly, the remaining studied biomarkers exhibited the decline order (albumin, ALB/GLB, total protein, creatinine and globulins) with the next data for every parameters [AUC = 0.641;  $P = 0.0676$ ; 95 % CI : 0.4937 to 0.7883 and SE : 0.07514], [AUC = 0.641 ;  $P = 0.0676$ ; 95 % CI : 0.4937 to 0.7883 and SE : 0.0676], [AUC = 0.6147 ;  $P = 0.0002$ ; 95% CI : 0.6543 to 0.8749 and SE : 0.07554], [AUC = 0.611 ;  $P = 0.1540$  ; 95% CI : 0.4621 to 0.7598 and SE: 0.07594], [AUC = 0.5369;  $P = 0.6320$ ; 95% CI: 0.4937 to 0.7883 and SE:0.0781] respectively, as shown in Table 3.

**Table 3: Area under Curve for Studied Parameters in Controls as compared to T2DM without and with COVID19.**

Parameter	AUC	Std. Error	95% confidence interval	P-value
Age	0.5491	0.0777	0.3968 to 0.7014	0.5281
FSG mg/ dL	1	0	1 to 1	< 0.0001
HbA1c %	1	0	1 to 1	<0.0001
Urea mg/ dL	0.6569	0.07324	0.5133 to 0.8004	0.0438
Creatinine mg/ dL	0.611	0.07594	0.4621 to 0.7598	0.1540
T. proteins g/ dL	0.6147	0.07554	0.4667 to 0.7628	0.1334
Albumin g/ dL	0.641	0.07514	0.4937 to 0.7883	0.0676
Globulins g/ dL	0.5369	0.0781	0.3839 to 0.69	0.6320

ALB/GLB	0.641	0.07514	0.4937 to 0.7883	0.0676
Periostin ng/mL	0.9764	0.01554	0.9459 to 1.007	<0.0001

## Discussion

Liver and pancreatic effects, endocrine organs impairment through the infection may cause metabolic abnormalities and glucose development in COVID-19 survivors [15]. Many severe disorders in glucose homeostasis can cause by COVID-19. Storm of inflammatory cytokine, in conjunction with the consequences of glucotoxicity in COVID-19; followed by increased oxidative stress [16]. COVID19 infection affect glucose homeostasis, which facilitating both dysfunction of pancreatic beta cells and insulin resistance [17]. In all previous studies unusual requirements to maintain glycemic control of insulin had been detected, along with dangerous of glucose fluctuations. High level of glucose has imposed a large interest in beta-cell invasion studies, the ability of SARS-CoV-2 to destroy islets and invade by its identified receptors specifically [18].

In this paper, serum concentration of periostin was high in both T2DM without and with COVID19. The level of periostin was higher mainly in patients with COVID19 as compared to cases without COVID19, ( $p < 0.0001$ ). We believe that the level of periostin may be used in the COVID19 severity diagnosis.

In the situation of stress and/or inflammation, cytokines release such as clusterin, IL-13 and IL-17 increases from cells with inflammation. This lead to periostin release from epithelial cells [19-21]. In this paper, although the level of periostin was establish to be great in two T2DM groups than in HCs. This may be because the regulation of periostin is affected not only by the expression of clusterin but similarly via IL-13 and IL-17 especially in COVID-19 patients [13]. We recommend additional studies to investigate the role of periostin in the progression of T2DM and its relationship to COVID-19 in diabetic patients.

Our study presented that severity related biomarkers (ferritin and D-dimer) and inflammatory were greater in T2DM group with COVID19. Furthermore, urea and creatinine demonstrating function of renal were worse in T2DM group with COVID19, which means greater risk of kidney damage. These results correspond with recent study, where T2DM patients with COVID19 had higher levels of ferritin and D-dimer as compared to [22]. Furthermore, renal damage was stated to be an acute complication of COVID-19 [23]. This supports our result of worse function of kidneys in the two T2DM groups.

Many mechanisms were assumed to clarify the bad results in hyperglycemic or diabetic patients with COVID-19 infection. First, high inflammation biomarkers such as D-dimer and ferritin are intensely related with infection of COVID-19 [24]. T2DM patients with COVID19 were

established to have greater serum levels of these biomarkers [22], Second, COVID-19 infection has been related with abnormal coagulation pattern and thrombosis risk [25].

The probable significance for increased levels of glucose helping to give the virus perfect environments to coat the spike protein with polysaccharides, that can give the virus' immune system additional capabilities to manipulate host cells and cannot be easily distinguished by antibodies, this need fixing many lines together across many sources of evidence to show how higher level of glucose may be participate in the disease complications, such as participating in the coagulation dysregulation, features of thrombotic and the immune response driving into storm of cytokines. COVID-19 management, an essential part of it lies in controlling the level of glucose in the blood [26].

Our results showed that serum levels of total proteins and albumin were lower in T2DM patients without and with COVID19 than in HCs and serum levels of these variables were lower in patients with COVID19 compared to patients without COVID19, these results were conflict with a recent study on the effect of albumin on Covid-19 patients [27]. As previously confirmed, the negative conversion rate of virus is highly related with admission low concentrations of blood albumin. Numerous mechanisms may be contributed to clarifying the vital role of albumin levels on the negative conversion rate of virus in COVID-19 infection such as albumin can play a main role in colloid osmotic pressure (COP) homeostasis. Regulating albumin levels may affect signaling of cells, vascular permeability, redox homeostasis and adhesion of neutrophil. It also has antithrombotic and anticoagulant properties. Therefore, albumin participates in several important vital functions such as; contribute to protection from inflammation and vascular endothelium [28]. Furthermore albumin has anti-inflammatory and immunomodulatory effects through cytokine production modulation, reaction with products of bacteria and antigen-presenting cell function modulation [29, 30].

This study has many limitations, such as; the ample size was small and was completely from an Iraqi people. Big randomized prospective studies are still required to state results reliability; also, we are still not sure, if a high concentration of periostin is a cause or merely parameter for T2DM.

**In conclusion**, the present study found that serum levels of periostin were significantly higher in T2DM patients without and with COVID19 than in HCs, and it is strongly related with FSG and HbA1c. Periostin may act as a scaffold and remodeling protein in inflammatory responses.

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## علاقة البيريوستين ببعض المتغيرات الحيوية في مرضى السكري من النوع الثاني غير المصابين والمصابين بمرض فيروس كورونا 19

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### الخلاصة:

مرض السكري من النوع الثاني هو مرض مزمن يتميز بارتفاع مستوى الكلوكرز في الدم والذي ينتج من قلة افراز الانسولين وخلل في وظائفه او الحاتين كلاهما. العديد من الدراسات اثبتت ان مرض السكري من الاسباب المهمة للاصابة بمرض فيروس كورونا 19. تم تصميم الدراسة الحالية للتحقق من علاقة البيريوستين مع بعض المتغيرات الحيوية في مرضى السكري من النوع الثاني غير المصابين والمصابين بمرض فيروس كورونا 19.

**المواد وطرق العمل:** تضمنت هذه الدراسة 56 مريضاً، 28 منهم كانوا غير مصابين بفيروس كورونا 19 وال 28 المتبقين كانوا مصابين بفيروس كورونا 19 تم جمعهم من مستشفى الفلوجة التعليمي، كما احتوت الدراسة على 28 شخصاً ظاهرياً يبدون اصحاء متطابقين مع المرضى من حيث العمر والجنس والخلفية العرقية كمجموعة سيطرة. المستوى المصلي للبيريوستين تم تقديره باستعمال تقنية الانزيم المرتبط المناعي الممتاز، البروتين الكلي والالبومين تم تقديرها بواسطة الطرق اللونية (طريقة بايوريت) اما بقية المتغيرات الحيوية فقدرت باستعمال الطرق الانزيمية اللونية.

**النتائج:** أظهرت النتائج ارتفاع المستوى المصلي للبيريوستين، اليوريا والكرياتين في مجموعتي المرضى مقارنة بالاصحاء ( $p < 0.001$ )، في حين اظهرت المستويات المصلية للبروتين الكلي و الالبومين انخفاض معنوي في مجموعتي المرضى مقارنة بمجموعة الاصحاء ( $p < 0.001$ ). البيريوستين اظهر علاقة ترابطية ايجابية معنوية مع مستوى سكر الصائم، الهيموكلوبين السكري، اليوريا، الكرياتينين، معامل كتلة الجسم ونسبة الالبومين الى الكلوبولين ( $p < 0.01$ ). تحليل المسافة تحت المنحني للمتغيرات المدروسة اظهر الترتيب التنازلي التالي: سكر الكلوكرز الصائم، الهيموكلوبين السكري (1)، البيريوستين (0.976)، اليوريا (0.6596)، الالبومين، نسبة الالبومين الى الكلوبولين (0.641)، البروتين الكلي (0.6147) و الكرياتينين، الكلوبولين (0.611).

**الاستنتاجات:** ان المستوى المصلي للبيريوستين يمكن استعماله كأداة تشخيصية ممتازة لتشخيص مرض السكري للمصابين وغير المصابين بمرض فيروس كورونا 19