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Research Paper

Evaluating the Predictive Significance of Biomarkers for Vascular Complications in Diabetic Patients

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ABSTRACT

The complications remain a leading cause of morbidity and mortality among individuals with diabetes, particularly type 2 diabetes (T2D). As the global burden of diabetes continues to rise, early identification of patients at risk for complications such as coronary artery disease, stroke, nephropathy, and retinopathy is increasingly critical. Biomarkers have emerged as valuable tools in predicting the onset and progression of these complications. Among these, natriuretic peptides (NPs), including atrial natriuretic peptide (ANP) and brain natriuretic peptide (BNP), have shown promise due to their roles in cardiovascular and renal function regulation. This review evaluates the current evidence supporting the predictive utility of NPs and other key biomarkers in vascular complication risk stratification among diabetic patients. Lower concentrations of ANP and BNP are associated with insulin resistance and increased cardiovascular risk, while elevated levels of Human Natriuretic Peptides (HNP) have been linked to enhanced insulin sensitivity and lower incidence of metabolic syndrome features. These associations suggest a potential mechanistic link between NP levels, metabolic dysfunction, and vascular damage.

Objective: To assess the predictive value of specific biomarkers—Urea, Creatinine, Blood Sugar (BS), Insulin, Uric Acid, Troponin, and Human Natriuretic Peptides (HNP)—in diabetic patients.

Design. This study included biomarkers for diabetic patients in the Diabetes and Endocrinology Department of Al-Sadr Teaching Hospital in Najaf Governorate. The analysis included 90 men and women (aged 40–75 years). Anthropometric measurements and blood pressure were recorded.

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KEYWORDS: atherosclerosis, atrial natriuretic peptide (ANP), Human Natriuretic Peptides (HNP), Insulin, nephropathy, troponin.)

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1. INTRODUCTION

In 2013, 382 million people worldwide suffered from diabetes, mainly type 2 diabetes, according to the International Diabetes Federation. Approximately 592 million people (a 55% rise from 2013) will have diabetes by 2035, up from 194 million in 2003, according to statistics.(1). Individuals who have type 2 diabetes are far more likely to experience microvascular (retinopathy and nephropathy) and macrovascular (coronary heart disease and stroke) problems.(2). About 40% of those with type 2 diabetes experience complications that may eventually result in death. Even in the wealthiest nations, the costs of comprehensive multifactorial interventions surpass the resources of healthcare systems, despite the fact that they can lower the incidence of complications. This is especially true given the growing number of diabetics who experience complications. Additionally, new research shows that type 2 diabetes is more common at younger ages and, more significantly, that early problems, including high blood pressure and kidney failure, are more likely to develop.(3, 4). One type of coronary artery disease that is a consequence of diabetes is acute coronary syndrome (ACS). Although ACS may not exhibit any symptoms, it frequently manifests as a myocardial infarction or unstable angina.(5). Diabetes patients, on the other hand, frequently have milder symptoms in terms of both intensity and type, and silent myocardial infarction is not identified until after the diagnosis. Globally, ACS is a major source of morbidity and mortality. It happens when a coronary artery suddenly becomes blocked. Depending on where and how severe the blockage is, it may result in myocardial infarction or unstable angina.(6) .

Atrial natriuretic peptide (ANP), brain natriuretic peptide (BNP), C-type natriuretic peptide (CNP), and dendroaspis natriuretic peptide (DNP) are all members of the family of peptides known as natriuretic peptides (NPs), which share similar sequences and conformations. (7). NPs have a unique genetic makeup and are important for maintaining renal function, circulation, and endocrine health. While pro-ANP is a 126-amino acid peptide that is stored in atrial myocytes, ANP is generated by the heart.(8). By lowering blood pressure and cell fibrosis in response to cardiac overload, pro-ANP protects cardiovascular and renal function.(9). Although it was initially discovered in the brain of a pig, BNP is primarily produced in the ventricle. According to earlier research, BNP predicts cardiac dysfunction in patients with suspected heart disease more accurately than other peptides.(10). Vascular tone modulation and the preservation of salt and water homeostasis are both significantly influenced by natriuretic peptides. NP levels are noticeably increased in certain pathological circumstances, such as acute ischemic stroke and heart failure. The three receptor subtypes that NPs interact with to produce their actions are the NP receptors (NPR)-A, NPR-B, and NPR-C. Guanosine triphosphate is changed into cyclic guanosine monophosphate by NPR-A and NPR-B. ANP, BNP, and DNP are bound by NPR-A, while CNP is modulated by NPR-B activation. It is well known that ANP is more active than BNP, particularly when it comes to lipolysis stimulation. According to certain research, NPR-A has a ten-fold higher affinity for binding ANP than it does for binding BNP(11). Other data, however, indicated that BNP is preferred by NP receptors over ANP. Further research should examine this dispute. NPC-C mediates NP activities in cardiovascular disorders in addition to its role in NP clearance. To clarify the precise functions of NPR-C in different pathophysiological conditions, more research is required.(12). Over the coming decades, it is anticipated that the prevalence of diabetes will rise worldwide. In comparison to the current age of onset, type 2 diabetes is predicted to develop at a significantly younger age. It's unclear what causes diabetes. Nonetheless, it is known to be linked to inherited traits, persistent low-grade inflammation, insulin resistance, and an unhealthy lifestyle. (13). Complications from diabetes may result in higher rates of morbidity and mortality as well as significant medical costs. Therefore, preventing diabetes's morbidity and mortality requires a better knowledge of the disease's pathophysiological course and complications. Patients with type 2 diabetes frequently exhibit insulin resistance. Numerous investigations revealed a strong correlation between NPs and insulin resistance. Low ANP levels raise the likelihood of insulin resistance, most likely by activating the renin-angiotensin system, according to prior research. (14). Nevertheless, hypertension or chronic heart failure raises serum insulin levels, indicating that ANP only prevents insulin breakdown in the liver, kidney, and other organs and has no effect on insulin secretion. (15). Higher serum NTproBNP levels were linked to higher insulin sensitivity at baseline and during the Diabetes Prevention Program (DPP) for type 2 diabetes, according to a clinical investigation. This relationship held regardless of therapy, body mass index, or waist circumference. Therefore, NT-proBNP could be a reliable measure of insulin sensitivity. (16) . Numerous epidemiological studies have shown that people with insulin resistance who go on to develop type 2 diabetes typically have lower levels of ANP and BNP. Further evidence for natriuretic peptides' involvement in the development of diabetes may come from their well-established ability to stimulate natriuresis, diuresis, and vasodilation. Insulin resistance may contribute to reduced nocturnal blood pressure drop, left ventricular hypertrophy, and elevated processing of plasma ANP and BNP in hypertensive individuals. Furthermore, the genesis of hypertension has been linked to insulin resistance and hyperinsulinemia. The underlying mechanism may be that insulin inhibits the natriuretic activity of ANP, while ANP dramatically increases urine salt excretion. (17)

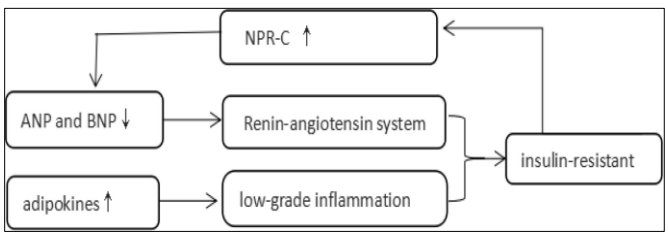

Fig. 1): Insulin resistance caused by persistent low-grade inflammation and renin-angiotensin system activation results in

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low levels of ANP and BNP and high levels of adipokines; insulin resistance inhibits circulating NPs by upregulating NPR-C expression. Atrial natriuretic peptide, or ANP; BNP: brain natriuretic peptide; NPR-C: natriuretic peptide receptor-C.

2. METHODS

Subjects and exclusion criteria

Ninety individuals participated in this study (62 with diabetes for more than ten years and 28 without diabetes). The sample included 52 males and 38 females from different areas of Najaf Governorate, Iraq. Age groups ranged from 45-75 years, and patients with acute infections, severe comorbidities, or recent surgeries were excluded. All participants were non-smokers. Samples were collected from December 12, 2024, to May 1, 2025, as shown in Table 1.

Table 1: Represents the number of patients with diabetes and the number of control patients.

		Sex		Total
		Male	Female	
Group	Patients	34	28	62
	Control	18	10	28
Total		52	38	90

Demographic Data and Medical Interview

At the Diabetes and Endocrinology Clinic at Al-Sadr Teaching Hospital, information was gathered in the morning. To gather data on education, smoking, employment, medical history, prior surgeries, current illnesses, and medication use, a thorough sociodemographic and medical interview was performed.

Anthropometric Measurements

Participants wore light clothing, and a stadiometer (Najaf, Iraq) was used to estimate body weight to the nearest 0.1 kg. A wall-mounted stadiometer (Salos) was used to measure height, and readings were rounded to the closest 0.1 cm. By dividing weight by height, the body mass index (BMI) was determined. A flexible tape measure was used to measure the waist circumference at the midpoint of the midaxillary line, which runs between the lower rib and the iliac crest. Samples were taken after participants had been seated for at least five minutes, and a power analysis was used to select the sample size in order to guarantee statistical significance.

Blood sampling protocol and laboratory analysis

Samples were drawn from the ulnar vein after a 13-hour fast. Samples were processed within 30 minutes of collection. Samples were separated by centrifugation at 2000 rpm for 15 minutes. Samples were stored at sub-zero temperatures for analysis. Glucose, urea, creatinine, and uric acid levels were measured by spectrophotometry, while troponin and human natriuretic peptides were measured by ELISA .

Statistical analysis

A statistical distribution splits a variable outcome into two types: nonparametric variables and normally distributed variables. The findings were presented as mean ± standard deviation for the normally distributed variable. The Pooled t-test was used to compare the measured parameters between the control and patient groups, as well as the subdivided groups. Pearson's correlation coefficients were computed to estimate the correlation between the parameters. When $p < 0.05$, the difference between groups is regarded as statistically significant. All statistical analyses were conducted using IBM USA and SPSS Statistics Base 26. While Microsoft Excel 2016 was used to organise the numbers (18).

3. RESULTS

Descriptive statistics

The number of males was 34, while the number of females was 28. The standard deviation (SD) of female length was 9.6, compared to 7.93 for males. Females also had a greater weight gain than males, and a slightly higher body mass index (BMI) for female than males, as shown in Table .2

Table 2: Shows a comparison between males and females in Patients with DM

Parameters	Male = 34	Female = 28	Total = 62	*p*
	Mean ± SD	Mean ± SD	Mean ± SD	
Age	57.68 ± 15.04	54.96 ±19.38	56.45 ± 17.04	0.537
Length (cm)	167.29 ± 7.93	166.32 ± 9.62	166.85 ± 8.68	0.664
Weight(kg)	70.62 ± 9.01	70.86 ± 10.59	70.73 ± 9.67	0.924
BMI	25.18 ± 2.34	25.55 ± 2.64	25.35 ± 2.46	0.568
Urea(mg/dl)	51.76 ± 17.63	43.39 ± 19.11	47.98 ± 18.64	0.078
Creatinine (mg/dl)	1.42 ± 0.57	1.41 ± 0.65	1.42 ± 0.60	0.968
Blood Sugar(mg/dl)	223.53 ± 94.79	200.39 ± 103.79	213.08 ± 98.81	0.363
Insulin(mg/dl)	167.44 ± 51.63	182.23 ± 56.90	174.12 ± 54.13	0.288
IR	90.34 ± 52.78	89.43 ± 57.08	89.93 ± 54.31	0.948
Uric Acid(mg/dl)	4.43 ± 1.20	4.54 ± 0.81	4.48 ± 1.04	0.68
Troponin	0.16 ± 0.08	0.14 ± 0.08	0.15 ± 0.08	0.23
HNP (pg./ml)	531.25 ± 82.83	505.31 ± 60.77	519.53 ± 74.27	0.173

BMI: Body Mass Index
IR: Insulin Resistance
HNP: Human Natriuretic Peptide

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Table 3 shows the comparison between the group of patients with DM and the control group, where the *P Value* was not significant as expected for age, weight, and body mass index, while urea, sugar, insulin, and insulin resistance were significant (p=0.001). Creatinine also had a significant value (p=0.003), while uric acid, troponin, and HNP were not significant, as the *P Value*s were (0.68, 0.23, 0.173), respectively.

Table 3: Shows a comparison between males and females in Patients with DM

Parameters	Patients 62	Control 28	Total 90	*P*
	Mean ± SD	Mean ± SD	Mean ± SD	
Age	56.45 ± 17.04	51.21 ± 13.18	54.82 ± 16.05	0.153
Length cm	166.85 ± 8.68	165.43 ± 9.61	166.41 ± 8.95	0.487
Weight kg	70.73 ± 9.67	69.82 ± 10.21	70.44 ± 9.79	0.687
BMI	25.35 ± 2.46	25.47 ± 2.79	25.39 ± 2.56	0.83
Urea (mg/dl)	47.98 ± 18.64	33.68 ± 8.90	43.53 ± 17.51	0.001
Creatinine (mg/dl)	1.42 ± 0.60	0.89 ± 1.02	1.25 ± 0.79	0.003
Blood Sugar(mg/dl)	213.08 ± 98.81	121.46 ± 29.78	184.58 ± 93.70	0.001
Insulin(mg/dl)	174.12 ± 54.13	114.08 ± 49.31	155.44 ± 59.39	0.001
IR	89.93 ± 54.31	34.81 ± 20.35	72.78 ± 52.97	0.001
Uric acid(mg/dl)	4.48 ± 1.04	4.33 ± 0.75	4.43 ± 0.96	0.483
Troponin(ng/l)	0.15 ± 0.08	0.19 ± 0.56	0.16 ± 0.31	0.653
HNP (pg./ml)	519.53 ± 74.27	488.84 ± 58.9	509.98 ± 70.98	0.039

BMI, Body Mass Index; IR, Insulin Resistance; HNP, Human Natriuretic Peptides; SD, Standard Deviation

Correlation analysis

A statistically significant positive relationship (r=0.329) between urea and creatinine (p-value=0.009), a statistically significant positive relationship (r=0.304) between urea and HNP (p-value=0.016), a statistically significant positive relationship (r=0.282) between urea and insulin (p-value=0.026), a Statistically significant positive relationship (r=0.746) between blood sugar and urea (p-value=0), a statistically significant positive relationship (r=0.746) (p-value=0), blood sugar and insulin resistance (r=0.736) (p-value=0), a statistically significant positive relationship (r=0.533) between insulin and insulin resistance (p-value=0.0), a statistically significant positive relationship (r=0.247) between insulin resistance and HNP, and a statistically significant positive relationship (r=0.247) between urea and insulin resistance (p-value=0.05).

Table 4: Analysis of the relationship between biomarkers and metabolic variables in DM patients

		Age	Length	Weight	BMI	Urea	Creatinine	Blood Sugar	Insulin	IR	Uric Acid	Troponin	HNP
Age	r	1	.252*	0.175	-0.006	.575**	0.102	.580**	-0.177	.387**	-0.11	-0.184	0.072
	P Value		0.048	0.175	0.965	0	0.429	0	0.17	**0.002**	0.396	0.152	0.579
Length	r	.252*	1	.722**	-0.039	0.055	-0.224	0.068	-.321*	-0.158	0.18	-0.072	0.232
	P Value	**0.048**		**0**	0.764	0.671	0.08	0.601	**0.011**	0.22	0.161	0.581	0.07
Weight	r	0.175	.722**	1	.660**	0.16	-0.117	0.178	-0.232	-0.033	0.204	-0.136	.272*
	P Value	0.175	**0**		**0**	0.213	0.363	0.166	0.069	0.801	0.111	0.293	**0.032**
BMI	r	-0.006	-0.039	.660**	1	0.178	0.067	0.196	0.012	0.132	0.097	-0.121	0.156
	P Value	0.965	0.764	0		0.165	0.607	0.127	0.926	0.305	0.452	0.349	0.226
Urea	r	.575**	0.055	0.16	0.178	1	.329**	.746**	-0.024	.631**	-0.205	-0.028	.304*
	P Value	**0**	0.671	0.213	0.165		**0.009**	**0**	0.855	**0**	0.111	0.828	**0.016**
Creatinine	r	0.102	-0.224	-0.117	0.067	.329**	1	0.202	.282*	.357**	-0.243	0.027	-0.012
	P Value	0.429	0.08	0.363	0.607	**0.009**		0.116	**0.026**	**0.004**	0.057	0.835	0.929
Blood Sugar	r	.580**	0.068	0.178	0.196	.746**	0.202	1	-0.129	.736**	-0.165	0.034	0.226
	P Value	0	0.601	0.166	0.127	**0**	0.116		0.317	**0**	0.2	0.791	0.077
Insulin	r	-0.177	-.321*	-0.232	0.012	-0.024	.282*	-0.129	1	.533**	0.045	-0.183	0.014
	P Value	0.17	**0.011**	0.069	0.926	0.855	**0.026**	0.317		**0**	0.731	0.155	0.915
IR	r	.387**	-0.158	-0.033	0.132	.631**	.357**	.736**	.533**	1	-0.129	-0.097	0.247
	P Value	**0.002**	0.22	0.801	0.305	**0**	**0.004**	**0**	**0**		0.317	0.454	**0.05**

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Pearson's correlation coefficient (r) is displayed. If a variable wasn't normally distributed, it was log-transformed before being used in the analysis. Ansate risk is used to identify log-Transformed variables (*). Very significant results (P ≤0.05) are bolded.

Fig 1: Receiver operating characteristic analysis of HNP and Insulin as Diagnostic Markers for DM

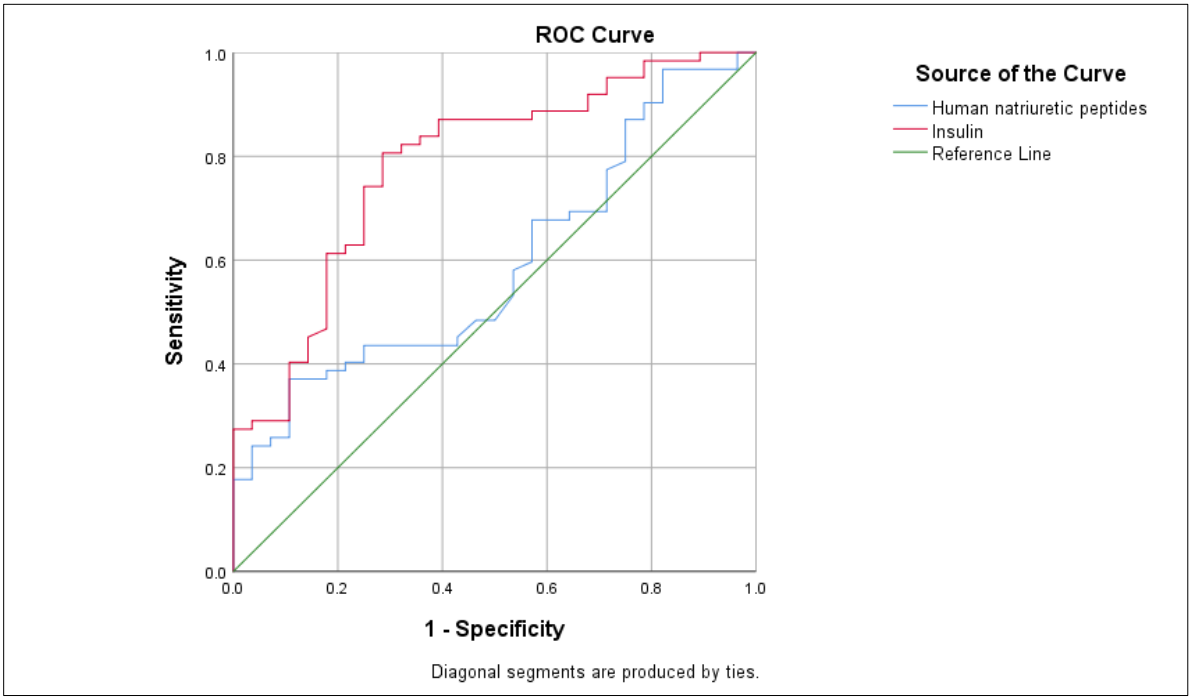

Table 5: Receiver operating characteristic analysis of HNP and Insulin as Diagnostic Markers for DM

Item	Cut off	Sensitivity	Specificity	AUC	95% CI of AUC	*P Value*
Human natriuretic peptides	502.73	58.1	46.4	0.593	0.471-0.715	0.159
Insulin	135.35	75.8	71.4	.784	0.682-0.887	0.0001

4. DISCUSSION

Evaluation of Biomarkers with DM and control groups

According to the study's findings, diabetics had higher levels of creatinine and urea. This is brought on by inadequate blood sugar regulation, which raises the risk of diabetic nephropathy in patients by raising serum urea levels. This is in line with earlier research findings that chronic kidney impairment is largely caused by elevated blood sugar. (19, 20).

In the current study, serum urea and creatinine levels were strongly correlated with the length of time and severity of diabetes. This result aligns with the notion that urea and serum creatinine are reliable indicators of glomerular filtration rate (GFR) (18). Compared to serum urea levels, serum creatinine is a more sensitive measure of renal function. The reason for this is that creatinine satisfies the majority of the criteria needed for an optimal filtration index. Males had higher amounts of creatinine and urea than females, according to this study. This could be because most men eat a high-protein diet, their muscles store creatinine as waste, and they have a lot of muscular mass. (21). The findings of the study are in line with prior research that has demonstrated that elevated levels of urea and plasma creatinine in diabetes people may be a sign of a kidney issue. In diabetic individuals, serum urea and creatinine levels can serve as helpful diagnostic markers and markers of kidney impairment. The morbidity and mortality linked to diabetic nephropathy can be significantly decreased by effectively controlling blood sugar levels, which can also prevent the development of this metabolic illness. The onset of nephropathy alterations is shown in the tendency for renal function test values to occur at higher reference limits in diabetes mellitus cases. (22). Renal function test estimation is now a viable supplement to the management and long-term treatment of diabetes mellitus because it is easy, dependable, affordable, and sensitive. (23).

The results of this study also showed a significant relationship between insulin and insulin resistance (p-value = 0.001) in diabetic patients compared to the control group. Insulin resistance is a condition in which the blood sugar response to insulin is lower than normal. Altered insulin sensitivity leads to several sets of responses. One set affects beta cells, leading to their rapid destruction and, consequently, the onset of complications of hyperglycemia. The other set generates a series of non-traditional cardiovascular risk factors that accelerate

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atherosclerosis; both sets of responses may affect other tissues, such as the nervous system, with a decrease in adiponectin release and an increase in free fatty acid and cytokine release. Increasing visceral adiposity is most likely the cause of insulin resistance (24).

This study also demonstrated a significant effect on HNPs when comparing diabetic patients with controls ($p = 0.039$). Increased HNPs in diabetic patients may be crucial in the development of diabetic retinopathy because they control vascular tone and restrict the proliferation of angiogenic cells. (25). In the early phases of diabetic retinopathy, HNPs are known to have anti-vascular permeability and anti-angiogenic actions. Additionally, HNPs are crucial in developing retinopathy's fibrotic and nonvascular consequences (proliferative diabetic retinopathy, or PDR). ANP levels in the vitreous humour are noticeably greater in patients with active neovascularisation than in those without active PDR. In DR, macular oedema typically causes vision loss to worsen. HNPs may decrease the function of the retinal pigment epithelium (RPE) barrier, which can lead to diabetic macular oedema, and lower the affinity of red blood cells for oxygen, which may cause cataract development. The N-terminal proBNP was linked to a risk of PDR in addition to nephropathy, neuropathy, and macrovascular disease in people with type 1 diabetes, indicating that NT-proBNP may be a useful prognostic indicator for diabetic sequelae. Therefore, HNPs are crucial for preserving the mature retina's vascular and neuronal integrity. (11). The results also showed a statistically insignificant difference in troponin levels in the diabetic group compared to the control group. This is due to several reasons, including that troponin takes at least 3-6 hours after heart damage to begin appearing in the blood. Furthermore, the effects of insulin resistance and chronic inflammation associated with diabetes may lead to changes in the heart muscle, but these changes are slow and cumulative and do not show sudden, large increases in troponin. Furthermore, most diabetic patients may suffer from chronic, gradual heart damage, such as diabetic cardiomyopathy, which results in a slight, persistent increase in troponin, rather than a sharp increase as occurs in a myocardial infarction (heart attack). This slight increase is not sufficient to demonstrate a significant difference when compared to those without diabetes.

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