Received: 23 November 2022 | Revised: 17 December 2022 | Accepted: 03 January 2023
DOI: 10.22034/ecc.2022.374021.1562
Journal of Medicinal and Pharmaceutical Chemistry Research
SAMI

FULL PAPER

Study of the level of signal-regulated kinase 5 (ERK5) in patients with coronary heart disease with and without diabetes mellitus type 2

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Diabetes mellitus (DM) is a complicated and varied metabolic disorder characterized by high blood glucose levels. Coronary heart disease (CHD), also known as ischemic heart disease or coronary artery disease, is a common term for the buildup of a waxy substance, called plaque, in the heart's arteries, resulting in the failure of coronary circulation to supply adequate blood circulation to cardiac muscle and surrounding tissue, which can lead to a myocardial infarction (MI). Diabetes mellitus (DM) and coronary heart disease are the most common non-communicable diseases in the world, causing morbidity and mortality due to microvascular and macrovascular complications due to the close relationship between diabetes and vascular complications.

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KEYWORDS

Coronary heart disease; diabetes; diabetes complication; ERK5; IL6.

Introduction

Atherosclerosis (AS), myocardial infarction (MI), heart failure (HF), ischemic stroke (IS), acute coronary syndrome (ACS), and coronary heart disease (CHD) are just a few of the illnesses that fall under the broad term of cardiovascular disease. Cardiovascular Diseases (CVDs) is the collective term for all heart and brain diseases connected to vascular diseases (CVD)[1]. Diabetes mellitus (DM) is a collection of metabolic syndrome recognized by rising in blood glucose, resulting impairment secretion of insulin. Diabetics lead to multiple metabolic disorders and irreversible damage to many tissues, and vital organs can be caused as a result for hyperglycemia if not controlled and managed well, such as ketoacidosis, foot diabetes (FD), diabetic retinopathy (DR), and neuropathy. Diabetes mellitus type 2 (T2DM) has characterized by reduction of cellular response to insulin (IR) or β cells distorted caused decrease in insulin level and its effect [2]. Patients with type 2 diabetes mellitus have more severe coronary atherosclerosis than those without diabetes. Endothelial dysfunction is aided by chronic hyperglycemia; hence, T2DM hastens the onset of serious coronary heart disease (CAD)-related problems [3]. Atherogenesis, development, and degeneration are all aided by the T2DM-induced modifications, which include dysregulation of vascular tone, increased permeability, inflammation, oxidative stress, and the pro-coagulant condition [4], as displayed in Figure 1.

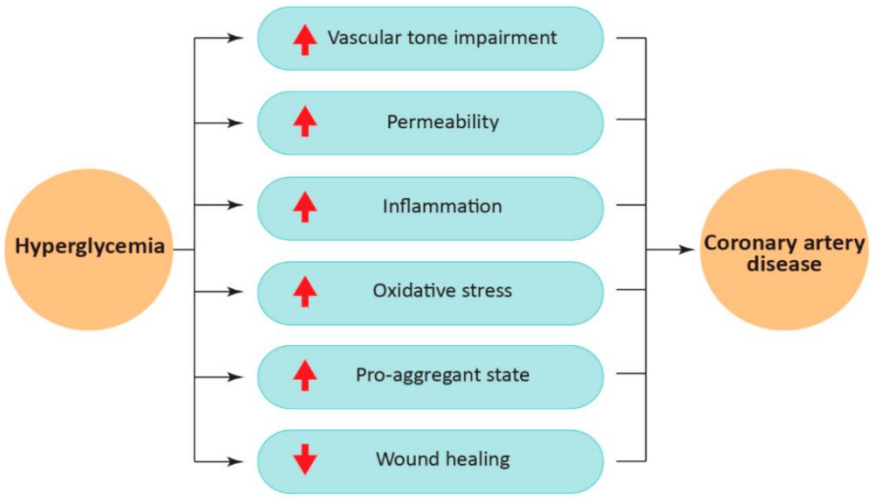

FIGURE 1 Hyperglycemia-induced mechanisms alter coronary artery disease etiology

The link between diabetes and coronary heart disease CHD is mediated by many patho-physiological processes. Numerous epidemiological studies back up the pathophysiological role of hyperglycemia because it directly affects endothelial function and the development and progression of atherosclerosis as demonstrated in Figure 2. However, there are also other pathophysiologic factors at play, including hyperinsulinemia, insulin resistance, and dyslipidemia. While dyslipidemia results in mitochondrial malfunction and subsequent cell death, hyperinsulinemia stimulates a number of inflammatory signaling pathways that promote the onset and progression of atherosclerosis. These mechanisms indicate similar routes for the development of both macro and microvascular problems, as well as heart and vasculature damage [5].

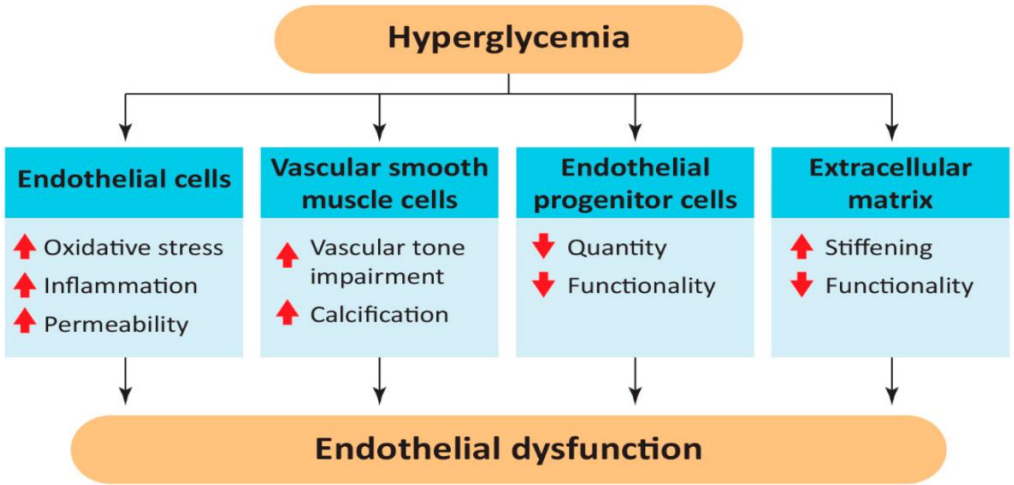

FIGURE 2 The effects of high glucose concentration on endothelial cells, vascular smooth muscle cells, circulating endothelial cells, and extracellular matrix that lead to endothelial dysfunction [5]

Interleukin-6 (IL-6) is a glycoprotein that has a weight of 26 kDa and is made up of 184 amino acids. In addition to have an effect on the immune system, IL-6 has a bearing on the neurological and cardiovascular systems [6]. IL-6 is an upstream cytokine that represents the inflammatory reaction, and it is also a possible biomarker for predicting the risk of CVDs [7]. The extracellular signal-regulated protein kinase 5 (ERK5) has 816 amino acids

and is predicted to have a molecular weight of 98 kilo Daltons (KD). It is a member of the family of mitogen-activated protein kinases (MAPKs) is also known as large mitogen-activated protein (MAP) kinase 1 (BMK1). Protein kinases are responsible for controlling many various biological functions, including cell proliferation, differentiation, and migration. They also govern the activation of the MAPK signaling pathway, which is phosphorylated in a sequential fashion. It is important in modulating the response of cells to mitogens, oxidative stress, and osmotic stress [8]. Extracellularsignal-regulated protein kinase (ERK5) has a unique and prolonged COOH-terminal that contains a transcriptional activation domain (TAD), the C-terminus ERK5 region at the end transcriptional activity is significantly inhibited by ERK5-SUMOylation at the NH2 terminal region [8,9]. In addition to protein localization, stability, and function, Small Ubiquitin-related Modifier (SUMO) posttranslational modification often controls protein complex assembly and disassembly. Besides being highly substoichiometric, SUMOylation produces stepper that lead to fresh protein interactions or conformational states [10] SUMO removal. SUMOylation targets ERK5 in diabetic hearts and heart disease. SUMOylation dependent ERK5 transcriptional repression is induced by ROS and high glucose levels [11]. ERK5 plays a critical role in numerous physiological activities, including on cell metabolism and, to some extent, in the control of lipid metabolism. This could explain the ERK5 significance in atherosclerosis and disease in which cholesterol levels form a risk factor, such as diabetes type 2 (DM2) [12,13]. Many evidences have studied the correlation between extracellular-signal-regulatedkinase 5 (ERK5) is required for proper gestational pancreatic beta cell proliferation [14].

This study aims to examine the comparison of extracellular signal-regulated kinase (ERK)5 activity in cardiovascular patients with and without type 2 diabetes mellitus.

Methods and materials

Participants

This investigation was done at Iraq's Ibn Al-Bitar Heart Hospital, International Research and Development Centre, and Educational Center during the period from December 2020 until April 2021. For 80 patients of both sexes with ages ranging from 35 to 65 years old with Ischemic heart disease (IHD) who have been divided into two groups:

1. Forty Patients with IHD and DMT2 (A Group).
2. Forty Patients with IHD and without DM (B Group).

Forty healthy individuals with the same age range (35-65 years old) comprise the control group (C Group).

All patients received clinical diagnoses from hospital physicians.

Blood sampling

Samples from the patients and control were collected after fasting 10-12 hours; from each participant subject (patient and control), 5-7 millilitres (mLs) of blood was collected the blood was divided into two parts:

1. Two mLs has been added to an ethylenee diamine tetraactic acid.

(EDTA) tube for the Glycated Hemoglobin (Hb1C) test.

2. The remaining blood was placed in a gel tube and centrifuged for 5 minutes for 3500 rpm and when the serum was separated, it was divided into several sections in several Eppendorff as in:

* Two hundred microliters (µls) for biochemictry measurements (Lipid profile, Glucose).

***** Two hundred µls for quantify, ERRK5, and IL6 (at 100 µl per measurement). These

Eppendorff tube stored in deep freeze (-20) °C.

This project consists of 120 individuals divided into three groups of 40 coronary artery disease patients (group A), 40 patients with diabetes with coronary heart disease (group B), and 40 control of healthy people with whom the pathological signs will be compared (group C).

Anthropometric examination

The weight was taken in kilograms (kg) and devided by the height in square meters, a body mass index (BMI) calculation was made (m^2), as indicated in the following equation: BMI = weight (kg)/Height (m^2).

Exclusion standards

Exclusion criteria for this trial were patients with Covid-19, cancer, alcohol assumption, smoker, hyperthyroidism, hypothyroidism, blood cancer, obesity, and kidney disease.

Ethical Considerations

The participants were informed of the research, their identities were kept confidential, and the results were unaffected by their involvement therefore the research was conducted according to our scientific committee's guidelines.

Methodology

Biochemical examination

Determination of fasting blood sugar Serum: Glucose is supplied as a liquid, ready-to-use, single reagent kit which contains reactive ingredients and inactive ingredients; supplied by Abbott / Canada. It was quantified with Architect C4000.

Determine Hb1C: HbA1C levels were estimated using a kit supplied by Alere Technologies/Osio, Norway and measured with an AfinionTM 2 Analyzer/Abbott Laboratories/Sweden as soon as possible after sample collection.

Determine lipid profile: High density cholesterol (HDLc), low density cholesterol (LDLc), and very low density cholesterol (VLDLc) are all components of serum total cholesterol (TC), triglycerides (TG), and (VLDL) are determined Abbott of Germany is the source of the by villas (TC, TG, and HDL). The lipid profile tests were carried out, making use of Spectrum auto-analyzer by Abbott with multiple channels (C4000, USA) instead of the Friedewald equation for LDL. = (TC-HDL-VLDL) and VLDL = TG/5 [15].

Determination of extracellular signal-regulated protein kinase5 (ERK5)

ERK5 level were determined according to the procedure developed by the mane facture (Melsin Medical Co. Limited / China) using double-antibody sandwich technique.

Measurement of IL6: IL6 level were determined according to the procedure developed by the mane facture (Shanghai Biological / China) using double-antibody sandwich technique.

Summary of the statistical tests

According to the study, results of high moral value ERK5 were obtained (p=0.000) and IL6 (p=0.000), lipid profile Cho, LDL(p=0.00), but LDL and TG ($P < 0.05$), HbA1c, and FBs test (p= 0.00).

Results and discussion

Table 1 presents the gender results for all groups. It was discovered that the percentage of men was larger than the percentage of women for each category divided from the other groups. Typically, men are diagnosed with coronary heart disease (CHD) years or even decades before women. This is related to the body's insufficient levels of estrogen [16], as explained later.

TABLE 1 Distribution of sample study according to sex in difference groups

Group	No.	Males No. (%)	Females No. (%)	P-value
Group A	40	21 (53.85%)	19 (46.15%)	0.257 NS
Group B	40	22 (55%)	18 (45%)	0.0079 **
Group C	40	21 (52.50%)	19 (47.50%)	0.257 NS
P-value	--	0.048 *	0.048 *	---

* (P≤0.05), ** (P≤0.01).

*Group A: Patients with IHD and DMT2.
*Group B: Patients with IHD and without DMT2.
*Group C: Healthy or control group.

Table 2 indicates Mean ± SE, Std., and Sig. of ERK5 and compares them for each study group's gender, as listed in the following table.

* A statistically significant difference was found between men and women in group A (non-diabetic heart population).

TABLE 2 Difference in Mean±SE, Std., Sig. of ERK5 between male and female for studied groups

	Group					
	A		B		C	
	Male	Female	Male	Female	Male	Female
Mean±SE	324.6667 ± 41.25030	324.6667 ± 41.25030	284.5000 ± 23.33260	238.6111 ± 31.33220	63.0476 ± 4.68659	86.8421 ± 11.00166
Std.Deviatn	189.03263	83.58859	109.43959	132.93127	21.47668	47.95514
Sig.	0.033		0.299		0.067	

*No significant difference was found between women and men in group B (diabetic heart population) and group C (control group)

TABLE 3 Mean Stander Error (Mean ± Std. Error), p-value, and mean Std., each criterion on the patient and control groups in the study

		Mean± Standard Error	Std.	P-value
Sex	A	1.46±0.081	0.505	0.976
	B	1.45±0.080	0.504	
	C	1.48±0.080	0.506	
BMI (Kg/m²)	A	26.3560±0.38009 a, b	2.40391	0.023
	B	24.8840±0.39342 a	2.48819	
	C	25.0820±0.43711 b	2.76450	
IL6 (ng/L)	A	168.54269±3.809574 ***b***	23.790784	0.000
	B	172.90307±5.626592 ***c***	35.585694	
	C	37.63300±3.880583 ***b, c***	24.542960	
FBS (mg/mL)	A	107.0769±2.57974 ***a***	16.11045	0.000
	B	187.8000±10.97557 ***a, c***	69.41558	

	C	94.4000±2.46483 ***c***	15.58895	
	A	5.9744±0.11817 ***a, b***	0.73796	
HbA1C%	B	9.2175±0.32869 ***a, c***	2.07882	0.000
	C	5.2863±0.09095 ***b, c***	0.57524	
	A	169.5128±16.26322 ***b***	101.56379	
TC (mg/dl)	B	147.8500±8.83289 ***c***	55.86410	0.000
	C	102.5250±4.99371 ***b, c***	31.58301	
	A	187.3077±8.12895 ***a, b***	50.76528	
TG (mg/dl)	B	163.3500±7.90103 ***a***	49.97053	0.010
	C	156.2000±5.98976 ***b***	37.88254	
	A	40.7692±1.60652 ***b***	10.03274	
HDLc (mg/dl)	B	38.5000±1.21740 c	7.69948	0.000
	C	47.8750±2.11290 ***b, c***	13.36315	
	A	111.8718±7.35207 ***b***	45.91364	
LDLc (mg/dl)	B	107.8750±6.80066 ***c***	43.01114	0.036
	C	90.1000±4.32046 ***b, c***	27.32501	
	A	33.9231±3.24118 ***b***	20.24116	
VLDL (mg/dl)	B	29.7250±1.92387 ***c***	12.16761	0.000
	C	20.7250±1.09368 ***b, c***	6.91705	
	A	310.3333±23.84077 ***a, b***	148.88557	
ERK5(pg/mL)	B	263.8500±19.16138 ***a, c***	121.18719	0.000
	C	74.3500±6.00572 ***b, c***	37.98350	

ANOVA test and spearman correlation were carried out to analysis gender, BMI, FBS, HbA1c, Chol , TG, LDL, HDL , VLDL, ERK5, and IL6 to patient with CHD, DMT2 patients, and control; in different models, as depicted in Tables 3, 4, and 5.

Anthropometric and biochemical results for two groups and control are listed in Table 3.

(Mean±SE) have high significant results for (IL6, FBS, HbA1C, Chol, HDL, VLDL, and ERK5), significant results for (Tri and LDL) and non-significant for (gander) of three groups at $p \leq 0.05$ level, as provided in Table 2.

A: Comparison between CHD group and T2DM with CHD group.

B: Comparison between CHD and control groups.

C: Comparison between T2DM with CHD group and control group.

Biochemical and anthropometric measurements of the participants are demonstrated in Table 3. The results showed significant differences for all parameters (except gender) between two groups and the control group. Men get coronary heart disease (CHD) faster than women. This is associated with estrogen deficiency. In other words, women acquire CVD on average ten years later than men, and the clinical presentation is typically unique [17].

Ovarian insufficiency, endothelial dysfunction, and a situation in which estrogen is an antiarteriosclerosis in midlife women [18,16]. In this study, there was no gender difference between patient groups because 80% of healthy and patient women were young. Especially in the first group (Group A) and the control group (Group C), there was no significant difference in either group, as shown in Table 3. However, there were statistically significant differences in BMI, IL6, FBS, Hb1C, Chol, Tri, HDL, LDL, VLDL, and ERK5 in the research groups.

Results for BMI, IL6, FBS, Hb1C, Chol, Tri, HDL, LDL, VLDL, and ERK5 all increased significantly in the group of patients with coronary artery disease (coronary heart disease) with and without DmT2, that was in

agreement with Hedayatnia *et al.* (2020) and Kumari *et al.* (2022) [19, 20].

According to Table 3, all the biometrics had indices, where IL-6 was a highly significant value that showed a clear increase Figure 6. IL6, a functional cytokine, found prognostic for numerous CVDs, including AS and CHD, that was consistent with the study conducte by Mickael Rosa *et al.* (2019) [21].

Long-term inflammation is a major cause of bad changes in the heart and the development of CHD. Circulatory factors that cause inflammation, like IL-6, were found to be higher in people with CHD [22].

An independent predictor of T2D is a high level of IL-6 in the blood, which is thought to be linked to inflammation, insulin resistance, and cell dysfunction. The results of FBS and Hb1C increased significantly in the group of patients with coronary artery disease (coronary heart disease) with and without T2DM because the rise in serum glucose levels (FBS) and Hb1C is caused by increases in glycogenolysis and gluconeogenesis, two endogenous glucose production routes [23].

Serum lipid profile levels was significantly higher in patients with CHD with and without DmT2 ones according Table 3,where CHD is caused by vascular epithelial tissue cell disease [24] this agreement with Yang X *et al.*(2019) [25]. Endothelial damage increases intimal permeability and leukocyte adhesion, facilitating thrombus formation and disease progression. Both ox-LDL and cholesterol damage artery intima, modify endothelial cell and leukocyte surface properties, and increase adhesion molecule expression [26].

Based on Table 3, the ERK5 data was significantly higher in patients with CHD with and without T2DM. There is a lot of evidence that ERK5 is a key part of keeping the cardiovascular system in balance, especially the vascular endothelium. The vascular endothelium is a layer of endothelial cells (ECs) that sits between the blood and the wall of the blood vessel. In the case of the CHD and T2DM, there is an inner lining in the damage, which is why the Erk5 level is increased [27].

TABLE 4 Pearson correlation for linear correlation measurement between two sets of data of CHD without T2DM patients

Correlations												
		Sex	**BMI.**	**IL6.**	**FBS.**	**HbA_{1C}**	**TC**	**Tri.**	**HDL c**	**LDL c**	**VLDL**	**ERK5**
Sex	**r.**	1	.082	.113	.1511	.195	-.246	.107	.277	.150	-.247	-.106
	p.		.613	.492	.36	.244	.130	.517	.088	.357	.130	.524
BMI	**r.**		1	-.240	-.047	-.02	-.028	.191	-.075	-.072	.191	-.261
	p.			.136	.774	.904	.865	.237	.646	.657	.238	.105
IL6	**r.**			1	.109	-.124	-.161	-.13	.263	-.106	-.155	.05
	p.				.511	.452	.327	.431	.105	.522	.345	.759
FBS	**r.**				1	.582**	-.003	-.196	-.132	-.161	-.003	-.211
	p.					.000	.984	.233	.422	.330	.988	.199
HbA_{1C}	**r.**					1	.110	.005	-.077	-.044	.109	-.266
	p.						.506	.974	.640	.789	.511	.100
TC	**r.**						1	.222	-.322*	-.143	1.00**	-.026
	p.							.175	.046	.392	.000	.878
Tri	**r.**							1	.288	.911**	.221	-.073
	p.								.075		.176	

HDLc	**r.**				.000		.655
	p.			1	.253 .121	-.322* .044	-.029 .866
DLc	**r.**					-.142 .393	-.063 .707
	p.				1		
LDL	**r.**						-.028 .872
	p.					1	
RK5	**r.**						1
	p.						

**. Significant correlation exists at 0.01 level

*. Significant correlation exists at 0.05 level.

Pearson's correlation coefficient of patients with CHD without DmT2 group can be seen in Table 4, which demonstrated:

* Gander, BMI, IL6, HbA1C, LDL, VLDL, and ERK5 were not significantly correlated. This disagrees with *J. J. Tsai (2020)* [28].

Because HB1C is a measure of levels of blood sugar over the last three months and is highly related to the chance of having DM. However, the patients in this case have CHD, not diabetes. Therefore, there was no statistically significant relationship between Hb1C and Gander in the CHD patient group [29].

* There is a significant negative correlation between HDL and VLDL that is in agreement with the study carried out by Koteliukh Mariia Yuriivna (2022) [29].

* A highly significant correlation was shown between FBS and Hb1C, VLDL, Chol., LDL, and Tri, that is in agreement with the study done by Sama Al-Shaheeb *et al.* (2022) [30].

Table 5 (Pearson correlation of patients with CHD with T2DM group) showed an association is found between:

* Gander and Chol (P=0.04). This could be because total cholesterol may be controlled by an ovary, which may explain the link between cholesterol and sex or because female hormones (estrogen) are also linked to cholesterol metabolism, that is consistent with the study conducted by Khamis RY *et al.* (2016) [31].

* BMI and FBS (P= 0.029); that is consistent with Nife Oudah N. *et al.* (2018) [32]. This is due to insulin resistance (increased resistance and decreased insulin sensitivity) leading to increased blood sugar, hyperglycemia, and higher BMI in those with T2DM [33].

* Chol. and Tri.(P=0.019), Chol. and LDL (p= 0.027), Chol. and VLDL (p= 0.000), Tri. and LDL (p= 0.000), and Tri. and VLDL (p=0.001) .

TABLE 5 Pearson correlation for linear correlation measurement of data of CHD with T2DM patients

Correlations											
		Sex	**BMI**	**IL6**	**FBS**	**HbA_{1C}**	**TC**	**Tri.**	**HDLc**	**LDLc**	**VLDL**	**ERK5**
Sex	**r.**	1	-.126	0.139	-.073	.007	.319*	-.013	.119	.216	.201	-.191
	p.		0.437	0.393	0.655	0.966	0.044	0.939	0.465	0.181	0.215	0.238
BMI	**r.**		1	-.073	.346*	.220	.095	.196	.005	.046	.081	.065
	p.			.656	.029	.173	.559	.227	.977	.780	.617	.691
IL6	**r.**			1	-.197	-.072	-.043	-.219	-.055	-.145	-.127	.049
	p.				.224	.657	.793	.175	.735	.372	.436	.762
FBS	**r.**				1	.293	-.006	-.178	-.016	-.232	.139	.098
	p					.066	.97	.271	.924	.150	.394	.549

	.							
HbA1c	**r.**	1	-.070	-.154	.135	.122	-.205	-.159
	P.		.668	.342	.405	.454	.204	.327
TC	**r.**		1	.368*	.074	.349*	.709**	-.117
	p.			.019	.650	.027	.000	.47
TG	**r.**			1	.139	.683**	.488**	-.139
	p.				.392	.000	.001	.393
HDLc	**r.**				1	.312	-.101	0.076
	p.					.05	.534	0.64
LDLc	**r.**					1	.094	-.195
	p.						.562	.228
VLDL.	**r.**						1	-.134
	p.							0.41
ERK5	**r.**							1
	p.							

*. Significant correlation exists at 0.05 level.

**. Significant correlation exists at 0.01 level

As depicted in Table 6, the relationship analysis for control parameters was as follow:

* The association between sex and (IL6, Tri, HDL, and LDL) showed a positive correlation with a higher significant value between (P =0.002 and P=0.009), while a negative correlation was shown between sex and (Chol and VLDL).

Estrogen is considered as an antilipidemic in women, which explains why women develop more atherosclerosis and heart disease (formed by fat accumulation) after menopause due to the loss of estrogen. That explains why the sex difference with lipids is related. That agrees with Zhu D. *et al.* (2019) [22]. Sex and ERK5 was shown moderate with (P < 0.05).

* The correlation between IL6 and other parameters was found to be significant (P 0.05) and positive for HDL and LDL. Chol (P=0.000) and VLDL (P=0.001) with (negative correlation).

* FBS was not significant with IL6, Chol, Tri, LDL, VLDL, and ERK5 except HBA1c (P= 0.001), higher significant and HDL (P< 0.05) was significant (negative correlation).

* Chol has a significant value with (Tri, HDL, LDL, and ERK5). However, Tri and HDL (P<0.05), LDL and ERK5 (P=0.005) (negative correlation, while it has no significant value with VLDL (P=0.000).

* Tri with HDL (P= 0.032) and LDL (P=0.00), it was significant and higher significant, respectively.

* HDL with VLDL (P= 0.047), it was significant with negative correlation.

* LDL with VLDL (P= 0.002), it was higher significant with negative correlation.

* VLDL has a significant value with ERK5 (P= 0.013).

TABLE 6 Pearson correlation for linear correlation measurement of parameters of control group

Correlations												
		SEX	BMI	IL6	FBS	HbA_{1C}	TC	TG	HDLc	LDLc	VLDL	ERK 5
Sex	r.	1	214	.340*	-.22	-.303	-.443**	.407**	.476**	.484**	-.409**	.317*
	p.		185	.032	173	.057	.004	.009	.002	.002	.009	.046
BMI	r.		1	-.016	040	-.136	.269	.033	.212	.327*	-.029	.224
	p.			.924	.809	.404	.093	.842	.189	.039	.858	.165
IL6	r.			1	-.009	-.216	-.568**	.259	.326*	.364*	-.514**	.169
	p.				.955	.180	.000	.107	.040	.021	.001	.298
FBS	r.				1	.493**	.025	-.034	-.312*	-.06	.001	.145
	p.					.001	.879	.834	.050	.712	.996	.371
HbA_1c	r.					1	-.007	-.143	-.249	-.046	-.018	.065
	p						.965	.379	.121	.776	.912	.691
TC	r.						1	-.323*	-.365*	-.440**	.951**	-.436**
	p.							.042	.021	.005	.000	.005
TG	r.							1	.340*	.688**	-.236	.280
	p								.032	.000	.142	.080
HDLc	r.								1	.300	-.316*	-.046
	p.									0.060	.047	.776
LDLc	r.									1	-.479**	.310
	p.										.002	.052
VLDL	r										1	-.390*
	p											.013
ERK5	r.											1
	p.											

*. A significant correlation exists at 0.05 level.

**. A significant correlation exists at 0.01 level.

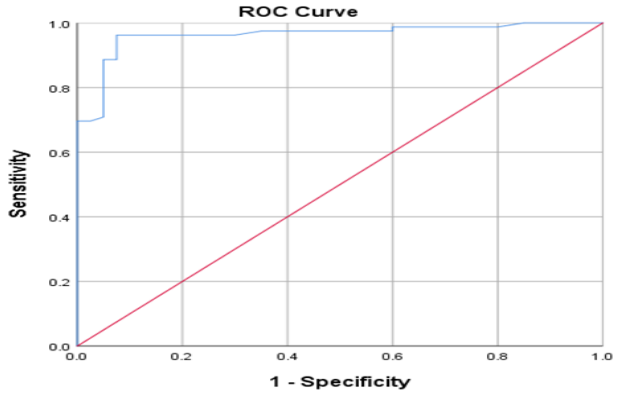

FIGURE 3 Roc relation between patients and control ERK5

According to the results obtained from the ROC relationship between patients and the control group (Figure 3), the results showed a strong association with ERK5 and the CHD

development. That's why ERK5 can be used for heart destruction.

TABLE 7 Area under the curve and cutoff value of ERK5 for total patients and control groups

Area	ERK5 Cutoff Value	Sensitivity	Specificity
0.963	108	96.25%	92.5%

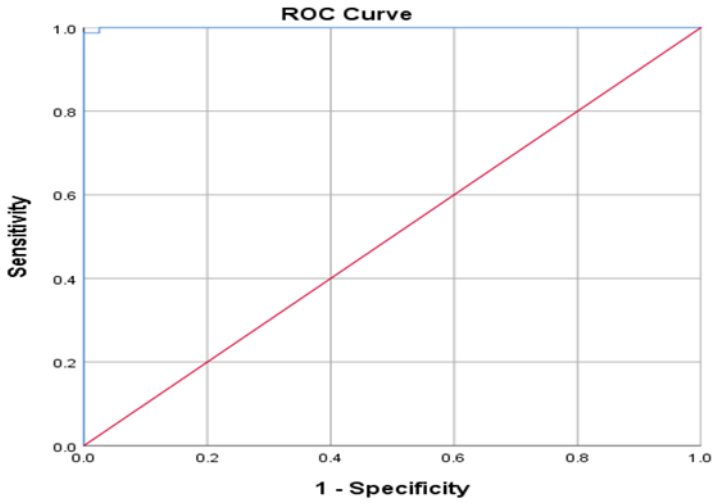

FIGURE 4 Roc relation between patients and control IL6

For ERK5, the results were AUC (area = 0.963 and sensitivity = 96.2%), as Table 7. This is an indication that the increase in these two measures is an indication of the development and occurrence of cardiovascular disease. Cardiovascular illnesses, such as coronary atherosclerosis, myocardial infarction, and stroke can be caused by anomalies in the ERK5 signaling pathway, which plays a vital role in the proliferation and differentiation of cardiac cells [28,34].

TABLE 8 Area under the curve and cutoff value of IL6 for total patients and control groups

Area	IL6 Cutoff Value	Sensitivity	Specificity
1.000	102.4	98.7%	97.5%

The results of IL6 according to AUC readings (area = 1 and sensitivity = 97.5%) showed a strong correlation to the presence of the disease in patients as Table 8.

Low-grade inflammation is known to play a major role in the development of many diseases, such as Type 2 diabetes and heart disease. IL-6 is one of the most important factors that cause inflammation, and that's why the association between heart disease and diabetes and Il6 was very strong [35].

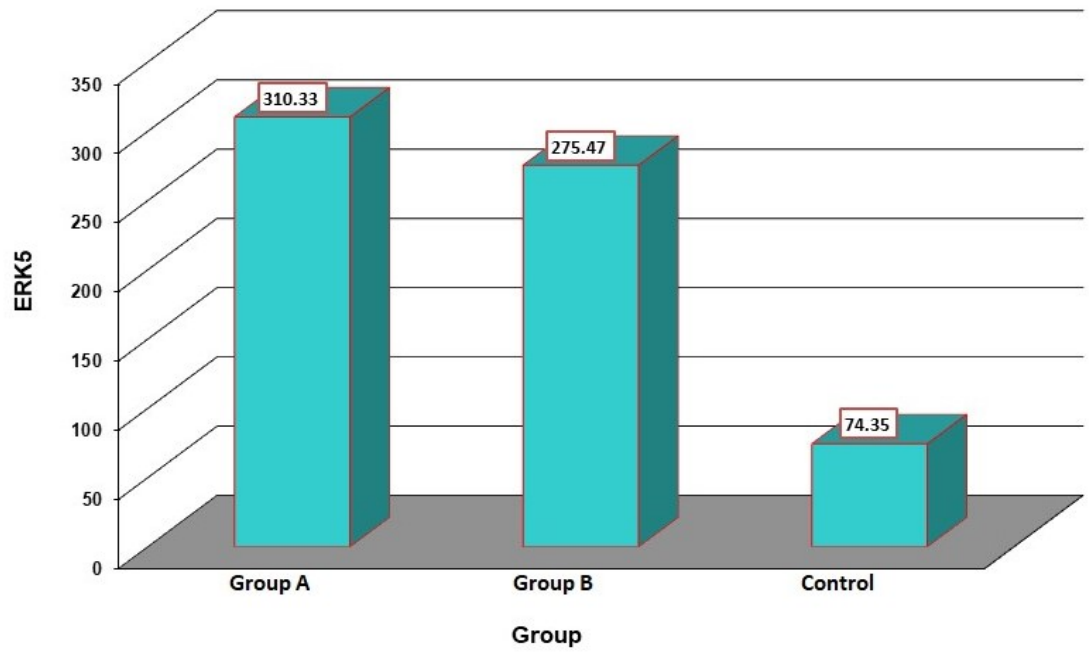

FIGURE 5 Comparison between difference groups in ERK5

ERK5 outcomes were significantly greater in patients with coronary heart disease and with and without T2DM compared with those without (Figure 5). There is a plenty of evidence to suggest that ERK5 is an important element in maintaining cardiovascular balance, particularly vascular endothelium. A vascular endothelium is a layer of endothelial cells (ECs) that lies between the blood and the wall of the blood artery. These cells are responsible for maintaining the lumen of blood vessels. In cases of coronary heart disorder and DmT2, damage has an internal lining, and this is the reason for a high Erk5 level [28].

Consequences of type 2 diabetes can be avoided by focusing on ERK5. Hyperglycemia suppressed ERK5 activation, which boosted ET-1, VEGF, and FN expression [29].

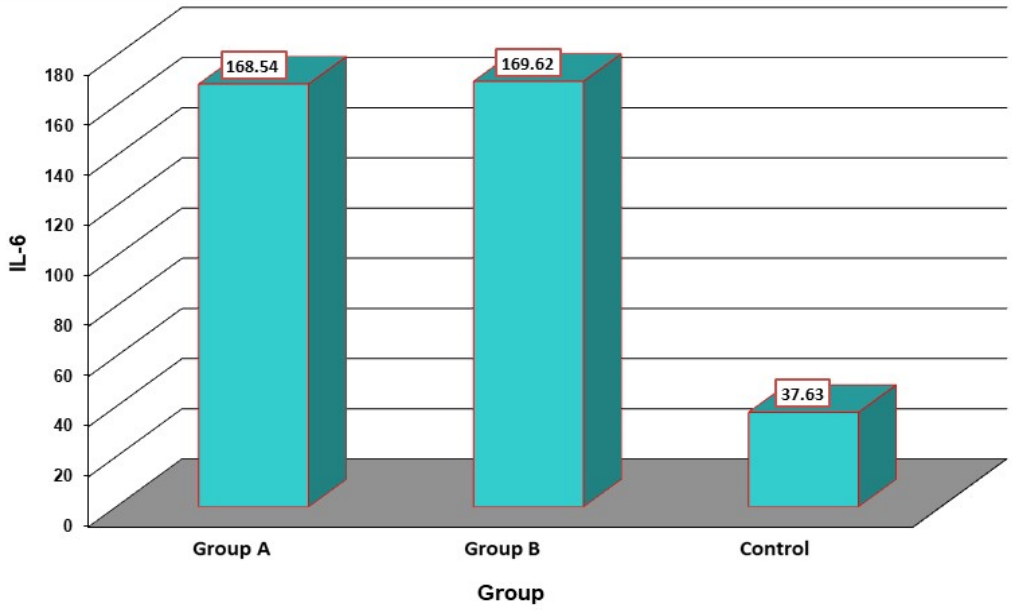

FIGURE 6 Comparison between difference groups in IL6

IL-6 is an interesting target in diabetes. IL-6 makes it easier for the liver and skeletal muscle to get rid of glucose when insulin is present, and treatment with IL-6 infusions (5 g/hour) has been shown to make it easier for humans to get rid of glucose when insulin is present. IL-6 is further released from the skeletal muscle when it is used. In addition, it has been revealed that IL-6 makes people make more insulin by making them make more glucagon-like peptide 1 [36]. Insulin resistance is increased by chronic inflammation, while tocilizumab's inhibition of IL-6 reduced insulin resistance. The elevated levels of IL-6 have found in people with T2D in multiple investigations, and they are probably associated to an abundance of adipose tissue. In such a tissue, macrophages produce IL-6, which encourages the oxidation of fatty acids and, in turn, lipolysis. It is possible that the extra IL-6 that macrophages in adipose tissue create could worsen insulin resistance and encourage gluconeogenesis in the level [37,38].

Conclusion

ERK5, particularly in the vascular endothelium, is an essential component in the homeostasis regulation process that keeps the cardiovascular system functioning normally. It is an important regulator of how heart disease and DM progress. It can be used to spot early signs of cardiometabolic disease and as a therapeutic target. When cardiovascular patients have the high levels of ERK5, this is a good sign that they will get CAD. Because the ERK5 level was significantly higher in people with CAD and DM, it can be used as a biomarker. The cytokine IL6 is a predictor of many CVDs, including AF and CAD. IL-6 is a primary cytokine that signals the inflammatory response. It could also be used to predict the CVDs risk.

Acknowledgements

The authors express their thanks and gratitude to all those who have helped them, in particular, Iraq's Ibn Al-Bitar Heart Hospital, International Research and Development Centre.

Conflict of Interest

The authors declare no conflict of interest.

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How to cite this article: Hibah Rajaa Taher*, Perry Habib Saifalla. Study of the level of signal-regulated kinase 5 (ERK5) in patients with coronary heart disease with and without diabetes mellitus type 2. *Journal of Medicinal and Pharmaceutical Chemistry Research*, 2023, 5(5), 425-440.