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Research Article

Study the Effects of the IL-1β -511 C>T Rs16944 SNP on the Incidence of Cardiomyopathy

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Abstract:

Background: The diseases that involve the myocardium directly and are not the result of hypertension, congenital, vascular, coronary, or pericardial abnormalities are called cardiomyopathy (CM). It is known that interleukin-1 (IL-1) family cytokines involve in inflammation and immune regulation and act as an important role in innate and adaptive immunity.

Aim: In this study, it was investigated the antioxidant capacity and their relation trace elements in cardiomyopathy CM patients and its correlation with IL-1β -511 C>T rs16944 SNP.

Methods: TAOC, and Cu-Zn SOD were determined by ELISA, and Cu and Zn were assessed by spectrophotometric method. IL-1β -511 C>T rs16944 SNP was assessed by PCR and DNA sequencing.

Results: The results were conducted that no statistically significant difference in age between CM and CONT groups (P>0.05). The results showed that was a statistically significant decrease in TAOC and SOD levels in the CM group compared to the CONT group (P<0.001). There was a statistically significant decrease in Cu and Zn levels in the CM group compared to the CONT group (P<0.001).

Conclusion: This study suggested that the essential role of antioxidant capacity and their trace elements affected by IL-1β -511 C>T rs16944 SNP as a risk factor for the incidence of CM in the Iraqi population.

Keywords: Cardiomyopathy, IL-1β, rs16944SNP, Antioxidant, Trace elements.

Introduction

The diseases that involve the myocardium directly and not the result of hypertension, congenital, vascular, coronary, or pericardial abnormalities are called cardiomyopathy (CM). The CM are diseases that involve the myocardium directly and not the result of hypertension, congenital, vascular, coronary, or pericardial abnormalities (1). The three major types of cardiomyopathy: dilated, hyperatrophic, and restrictive (2). Intense myocardial dead tissue (AMI) is characterized as myocardial corruption because of an interference of coronary blood supply (which decline the conveyance of oxygen and supplements) as well as because of expanded metabolic interest (3). A break in the stockpile of myocardial oxygen and supplements happens when a blood clot is superimposed on a ulcerated or temperamental atherosclerotic plaque and results in coronary impediment. Ischemic myocardial cell injury can be either reversible or irreversible (4). Dazzling and hibernation of the myocardium are types of reversible myocardial harm frequently connected with more limited season of ischemia or with reperfusion injury. Whenever the myocardium is presented to longer times of ischemia (20-30 min) histological cell demise happens (5). Complete rot of myocardial cells in danger expects somewhere around 2-4 h of ischemia, or longer, contingent upon the presence of security dissemination to the ischemic zone, constant or discontinuous coronary conduit impediment, the responsiveness of the myocytes to ischemia, preconditioning and individual interest for oxygen and supplements (6). Actual interruption of the atherosclerotic plaque usually causes blood vessel apoplexy by permitting blood coagulant variables to contact thrombogenic collagen found in the blood vessel extracellular lattice and tissue factor created by macrophage-inferred froth cells in the lipid center of sores. As such, locales of plaque crack structure the nidus for thrombi (7). The typical vein divider has a few fibrinolytic or antithrombotic instruments that will generally oppose (8). Cancer prevention agents are a complex gathering of compounds for fix of harmed DNA , harmed protein, oxidized lipids and peroxides and furthermore to stop chain engendering of peroxyl lipid revolutionary. These catalysts fix the harm to biomolecules and reconstitute the harmed cell layer, for example lipase , proteases, DNA fix catalysts, transferase ,methionine sulphoxide reductase, and so on (9). It is known that interleukin-1 (IL-1) family cytokines involve in infammation and immune-regulation and act as an important

role in innate and adaptive immunity (10). IL1 family genes occupy nearly 400Kb including three well-defined genes IL1B, IL1A and IL1RN and their products IL1α and IL1β are agonists, while IL1RN is antagonist (11). The aim of this study is assessment of antioxidant capacity and their relation Trace elements in Iraqi cardiomyopathy patients.

Materials and methods

Study design

All samples were collected from coronary care unit (CCU) in Merjan teaching hospital. This study included 75 patients, 45 males and 30 females and range of age was 42-79 years. Control group included 50 subjects 30 male and 20 females and the range of age was 40-73. The patients were diagnosed as (ST-segment elevation myocardial infarction) depending on positive troponin I tests and ECG in addition to clinical features of CM. All the patients were fasting when blood samples were taken.

Determination of Total Antioxidant Capacity TAOC

The aantioxidants were suggested that play an important role in decrease the levels of oxidative stress and the procedure was done depending on ELISA kit, as shown in figure 1.

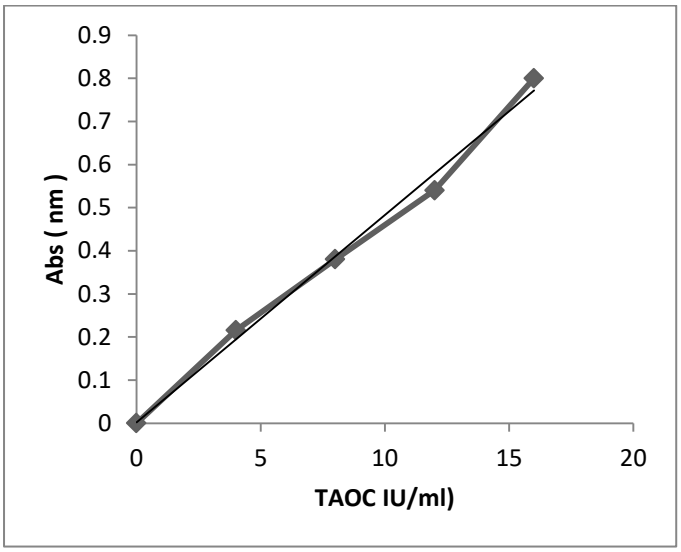

Fig. 1: TAOC standard curve

Superoxide Dismutase Activity SOD:

Cu - Zn SOD not entirely settled by utilize a straightforward and quick strategy, in view of the capacity of the chemical to restrain the autioxidation of pyrogallol . The autoxidation of pyrogallol within the sight of EDTA in the pH 8.2 is half. Assimilation is perused at the frequency of 420 nm against Tris-EDTA Buffer at zero time and following 1 moment of the expansion of pyrogallol .

$$\% \text{ Inhibition of pyrogallol autoxidation} = \frac{?A \text{ test}}{?A \text{ control}} \times 100\%$$

$$\text{Cu - Zn SOD activity } \frac{U}{ml} = \frac{\% \text{ Inhibition of pyrogallol autoxidation}}{50\%}$$

Determination of serum copper and zinc

Spectrophotometric method was used to determine the trace elements (Cu and Zn) in serum samples depending on protocol provided by manufacture.

IL-1β -511 C>T rs16944 SNP genotyping analysis

The DNA were extracted from blood samples and carried out using the genomic purification kit followed by using 2µl of each sample to evaluate DNA concentration and purity by using analytikgena^{R} Nanodrop Spectrophotometer, then the DNA integrity detection 2% agarose gel was used. The IL-1β genotyping was was considered as a candidate gene in which there were SNPs associated with CM. F and R primers were used its sequences, F:5`-TGGCATTGATCTGGTTCATC-3`, R:5`- GTTTAGGAATCT TCCCACTT-3`.

PCR were performed to estimation of IL-1β -511 C>T rs16944 SNP genotyping , The PCR reaction mixture was subjected to an initial denaturing step of 4 min at 95 °C, then 40 cycles of denaturing for 45 sec at 95 °C, annealing for 30 sec at 60.7 °C for, extension for 60 sec at 72 °C, and a final extension step of 10 min at 72 °C followed by the examination of PCR product 2% agarose gel, as shown in figure 2:

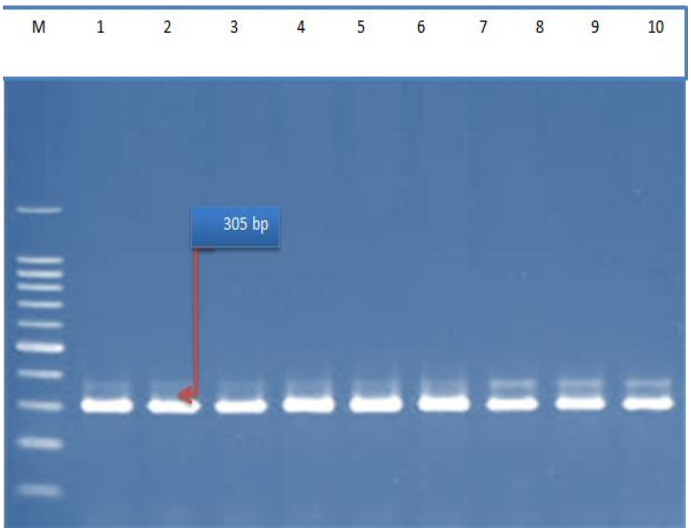

Fig.(2): IL-1β -511 C>T rs16944 SNP PCR product (305 bp)

PCR products(DNA samples) from CM and CONT groups were subjected to sequencing in bioneer ® company (South Korea) to determine the GG, AG, and AA genotypes of IL-1β -511 C>T rs16944 SNP as shown in figure 3:

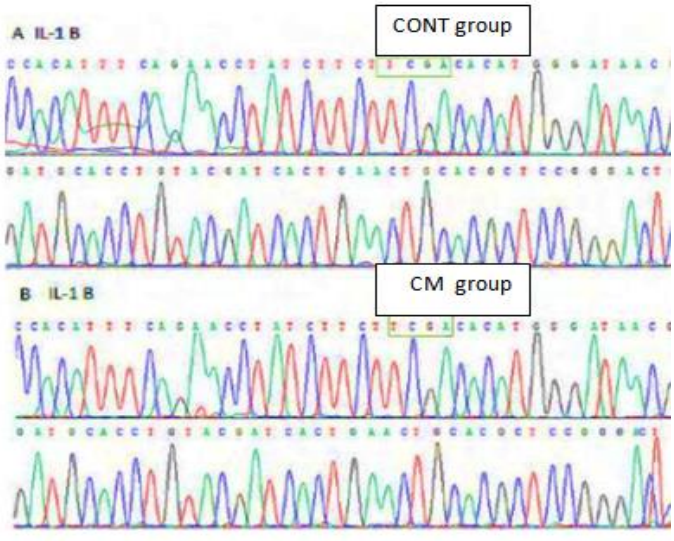

Fig. (3): DNA sequencing of CM and CONT samples

Results

There was no statistical significant difference in age between CM and CONT groups (P>0.05) , as shown in table (1) .

Table (1): Clinical characteristics of patients CM and control CONT group depending on age.

groups	Number	Age (years) Mean ±SD	P value
CONT	50	60.44± 10.1	P>0.05
CM	75	61.74 ± 11.8	

The results showing that was a statistical significant decrease in TAOC levels in CM group compared to CONT group (P<0.001), as shown in table (2).

Table (2): TAOC levels in CM and CONT groups

Groups	Number	TAOC Mean ±SD IU/ml	P value
CONT	50	6.322 ± 1 .05	P<0.001
CM	75	2.099 ± 0 .765	

The results suggested that was a statistical significant decrease in SOD levels in CM group compared to CONT group (P<0.001) as shown in table (3).

Table (3): SOD levels in CM and CONT groups

Groups	Number	SOD Mean ±SD IU/ml	P value
CONT	50	34.343± 3.55	P<0.001
CM	75	11.123 ± 1 .912	

There was a statistical significant decrease in Cu and Zn levels in CM group compared to CONT group (P<0.001) as shown in table (4).

Table(4): Serum levels of Cu and Zn in CM and CONT groups

Sample	Number	Zn Mean ±SD mg/dl	Cu Mean ±SD mg/dl	P-value
CONT	50	68.77 ± 4.79	175.47 ± 14.21	P<0.001
CM	75	34.65 ± 2.49	98.65 ± 10.08	

The frequency of the genotypes and alleles of IL-1β -511 C>T rs16944 SNP in both CM and CONT groups were estimated according to Hardy-Weinberg equilibrium (HWE) that listed in table 5:

Table (5): Comparisons of genotype distribution, allele frequency, and genetic models between CM and CONT groups

SNP ID	GENOTYPES	CM N= 75(%)	CONT N=50(%)	OR(CI,95%)	P-VALUE
rs16944	AA	45(60)	20(40)	1(Reference)	-
	AG	20(27)	24(48)	1.9(0.6-2.7)	0.012*
	GG	10(13)	6(12)	3.8(0.9-4.9)	0.001**
Allele	A vs. G	45/30	25/25	4.5(1.5-5.8)	0.001**
Dominant	GG+AG vs. AA	30/45	25/25	5.7(1.7-4.6)	0.001**
Recessive	GG vs. AA+AG	10/65	6/44	4.8(1.3-3.9)	0.001**
Homozygous	GG vs. AA	10/45	6/25	3.9(1.8-4.2)	0.001**
Heterozygous	AG vs. AA	20/45	19/25	4.4(1.3-4.9)	0.001**

* (P<0.05), **(P<0.0001)

Discussion

The two category the first is primary that include of genetic, mixed, or acquired and the secondary categories, which result in varied phenotypes including dilated, hypertrophic, and restrictive patterns has been called cardiomyopathy CM. The hypertrophic cardiomyopathy is the most common primary CM and can cause exertional dyspnea, presyncope, atypical chest pain, heart failure, and sudden cardiac death (12). There was no statistical significant difference in age between CM and CONT groups (P>0.05). Enlarged cardiomyopathy can be hereditary or obtained and regularly gives exemplary side effects of cardiovascular breakdown with diminished launch portion. Prohibitive cardiomyopathy is significantly less normal and frequently connected with foundational infection (13). Cell reinforcements are acomplex gathering of compounds for fix of harmed DNA , harmed protein, oxidized lipids and peroxides and furthermore to stop chain proliferation of peroxyl lipid revolutionary. These chemicals fix the harm to biomolecules and reconstitute the harmed cell film, for example lipase , proteases, DNA fix chemicals, transferase ,methionine sulphoxide reductase, and so on((14). Risk factors for cardiovascular infection, for example, hypertension, hypercholesterolemia, diabetes mellitus, and cigarette smoking, as well as cardiovascular sickness itself, are completely connected with huge expansions in ROS in the vascular divider, a circumstance that at last finishes with oxidative pressure. In the condition of oxidative pressure, enzymatic creation of ROS surpasses the accessible cell reinforcement safeguard frameworks (15). The outcomes showing that was a factual huge reduction in TAOC levels in CM bunch contrasted with CONT bunch (P<0.001). Cancer prevention agents can be little natural particles, for example, ascorbate and urate, or compounds, for example, superoxide dismutase. Natural cell reinforcements can be either lipid

dissolvable (vitamin E) or water solvent, for example, glutathione (GSH), ascorbate, and urate (16). Vitamin E is certifiably not a solitary natural particle yet alludes to somewhere around eight normally happening compounds, four tocopherols (alpha-, beta-, gamma-and - delta), and four tocotrienols (alpha-, beta-, gamma-and - delta) which are all lipid solvent and related with lipid-protein buildings, for example, biomembranes and lipoproteins (17). There was a factual critical lessening in Cu and Zn levels in CM bunch contrasted with CONT bunch (P<0.001). Minor components assume a significant part in the construction of proteins, compounds and complex carbs to take an interest in biochemical responses. Fundamental minor components are associated with various metabolic exercises, including neuro conduction, transport, excretory cycles and filling in as cofactors for proteins (18). The C>T -511 SNP rs16944 was significantly different between CM and CONT group as the frequency of GG genotype was higher in CM than CONT (60% vs. 40%). As well as the G allele frequency (65% in CM, 35% in CONT group) and this results were approved its consideration as a risk factor to CM incidence in Iraqi population. This results agreement with finding of other studies (19-22). In conclusion, this study revaluated the essential role of antioxidant capacity and their trace elements and IL-1β -511 C>T rs16944 SNP in the incidence of CM.

Conclusion:

Deficiency in antioxidant capacity agents such as enzymes and its trace elements with IL-1β -511 C>T rs16944 SNP suggested that risk factor for the incidence of CM in Iraqi population.

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Conflict of interest

Non

References

1. Mohan S.B , Parker M . , Wehbi M., Doglass P. Idiopathic dilated cardiomyopathy : A common but mystifying cause of heart failure. Cleveland clinical journal of medicine 2002 : 69 (6) : 481-87.
2. Wynne J , Braunwald E. The cardiomyopathy and myocarditis. Principle of Internal Medicine 1999 , 2 : 1359-0.
3. Japp A.G., Gulati A., Cook S.A., Cowie M.R., Prasad S.K. The Diagnosis and Evaluation of Dilated Cardiomyopathy. *J. Am. Coll. Cardiol.* 2016;67:2996–3010.
4. Laonigro I., Correale M., di Biase M., Altomare E. Alcohol abuse and heart failure. *Eur. J. Heart Fail.* 2009;11:453–462.
5. Schäfer S., De Marvao A., Adami E., Fiedler L., Ng B., Khin E., Rackham O., van Heesch S., Pua C.J., Kui M., et al. Titin-truncating variants affect heart function in disease cohorts and the general population. *Nat. Genet.* 2017;49:46–53.
6. Ware J., Li J., Mazaika E., Yasso C.M., DeSouza T., Cappola T.P., Tsai E.J., Hilfiker-Kleiner D., Kamiya C.A., Mazzarotto F., et al. Shared genetic predisposition in peripartum and dilated cardiomyopathies. *N. Engl. J. Med.* 2016;374:233–241.
7. Taylor M.R., Fain P.R., Sinagra G., Robinson M.L., Robertson A.D., Carniel E., di Lenarda A., Bohlmeyer T.J., Ferguson D.A., Brodsky G.L., et al. Natural history of dilated cardiomyopathy due to lamin A/C gene mutations. *J. Am. Coll Cardiol.* 2003;41:771–780.
8. Lakdawala N.K., Winterfield J.R., Funke B.H. Dilated cardiomyopathy. *Circ. Arrhythm. Electrophysiol.* 2013;6:228–237.
9. Phaniendra A, Jestadi DB, Periyasamy L. Free radicals: properties, sources, targets, and their implication in various diseases. Indian J Clin Biochem. 2015 Jan;30(1):11-26.
10. Kornman, K.S. (2006). Interleukin 1 genetics, inflammatory mechanisms, and nutrigenetic opportunities to modulate diseases of aging. The American Journal of Clinical Nutrition, 83(2): 475-483.
11. Nicklin, M.J.H.; Barton, J.L.; Nguyen, M.; FitzGerald, M.G.; Duff, G.W. and Kornman, K. (2002). A Sequence-Based Map of the Nine Genes of the Human Interleukin-1 Cluster. Genomics Journal, 79(5): 718-725.
12. Maron M.S., Maron B.J. Clinical Impact of Contemporary Cardiovascular Magnetic Resonance Imaging in Hypertrophic Cardiomyopathy. *Circulation.* 2015;132:292–298.
13. MacIntyre C., Lakdawala N.K. Management of Atrial Fibrillation in Hypertrophic Cardiomyopathy. *Circulation.* 2016;133:1901–1905.
14. Sandalio LM, Rodríguez-Serrano M, Romero-Puertas MC, del Río LA. Role of peroxisomes as a source of reactive oxygen species (ROS) signaling molecules. Subcell Biochem. 2013;69:231-55.
15. Stone WL, Krishnan K, Campbell SE, Palau VE. The role of antioxidants and pro-oxidants in colon cancer. World J Gastrointest Oncol. 2014 Mar 15;6(3):55-66.
16. Swain N., Ray J. G. Altered trace element level and antioxidant activity in whole blood of oral leukoplakia and cancer patients in comparison with healthy controls. *International Journal of Oral & Maxillofacial Pathology.* 2011;2(2):2–6.
17. World Health Organization. *Trace Elements in Human Nutrition and Health.* Geneva, Switzerland: World Health Organization; 1996.
18. Mulware S. J. Trace elements and carcinogenicity: a subject in review. *3 Biotech.* 2013;3(2):85–96.
19. Ali SH, AL-Azawi RSA, Kzar HH. Study the IL6 (C174G) Promoter SNP and Correlation with Physiological Growth Hormone and TNFA levels in Iraqi Subjects with Psoriasis. *SRP*. 2020; 11(9): 272-276.
20. Kzar, Hamzah H.; Al-Gazally, Moaed E.; Wtwt, Moshtak A. Everolimus loaded NPs with FOL targeting:

preparation, characterization and study of its cytotoxicity action on MCF-7 breast cancer cell lines. JJPS ; 2022, 15 (1), p25-39.

21. Kzar,H,H.,Wtwt.M.A.,Al-Gazally ,M.E. Study the glucose transport, angiogenesis and apoptosis behavioral through chemotherapy treatment according to receptors status in women with breast cancer. Indian Journal of Forensic Medicine and Toxicology,2020,14,(3),pp.2555-2559.
22. Al-Gazally,M.E., Alaa S. Al-Awad, Hamzah H. Kzar. Study Some of the Risk Factors on Total Antioxidant Capacity in Iraqi Patients with Sporadic Colorectal Cancer. Journal of Global Pharma Technology,2018, 10(3), pp:48-55.