Unstable Angina Biomarkers and Their Main Risk Factors in Iraqi Patients

Ammal E. Ibrahim[*]**, Hiba A. Hasan**[**]**, Hedef D. El-Yassin**[***]
*Department of Chemistry, College of science, Al-Nahrain Uniersity,
**Department of Clinical Laboratory Science, College of Pharmacy, Al-Mustansiriyah
University.***Department of Physiological Chemistry, College of Medicine, University of
Baghdad.

: ألخلاصة

تهدف هذه الدراسة لتوضيح دور مستوى هرمون اللبتين والدهون عند المرضى العراقيين المصابين بالذبحة الصدرية غير المستقرة و كذلك لتحديد عوامل الخطورة الاساسية التي تتسبب في ارتفاعهما.

تشتمل هذه الدراسة على (37) مريض مصاب بالذبحة الصدرية غير المستقرة و (20) من الأشخاص الأصحاء المتقاربين بكل من العمر والجنس وكتلة الجسم.

التحليل الإحصائي المستخدم في هذه الدراسة هو اختبار t, اختبار الارتداد الخطي واختبار العلاقة. تعتبر الزيادة معنوية إذا كانت قيمة p<0.05.

طريقة جمع العينات كانت بواسطة طريقة الانتقاء المريح. أظهرت النتائج الاساسية لهذه الدراسة هي إن اللبتين و مستوى الدهون وخصوصا LDL, TG, TC و VLDL لكن ليس HDL هي العوامل البيولوجية الرئيسية في حدوث ألذبحة ألصدرية غير المستقرة المستقبلية عند المرضى العراقيين.

Abstract:

The presented study focuses on the main role of leptin and lipid profile level in Iraqi patients with unstable angina to indicate the main risk factors that play a role with their elevation.

This is a case control study conducted on (37) unstable angina patients and (20) healthy control who were closely similar by age, gender and BMI.

The main statistical analysis used was student *t* test, linear regression test and correlation test. Significance was set at $P < 0.05$. Sampling method used for this study was convenience sampling method.

The main results of this study showed that leptin and lipid profile specifically LDL, TG, TC and VLDL are the main biomarker for the future incidence of unstable angina in Iraqi patients. Doctors should focus on both leptin level and lipid profile level as biomarkers for future incidence of unstable angina patients.

Key- Words: Leptin, Lipid profile, Unstable Angina, Biomarker.

Introduction:

Leptin, a recently discovered obesity gene product, is primarily involved in the regulation of food intake and energy expenditure [1].
In human obesity, serum leptin concentrations are directly proportional to body fat mass and lipid profile, suggesting leptin resistance, rather than leptin deficiency, is the pathogenetic mechanism in human obesity [2]. Moreover recent observations suggest that leptin has a much broader biological role other than regulation of body weight and energy metabolism [1]. Since it has been shown that leptin has angiogenic activity that promotes vascular smooth muscle cell migration and proliferation [3]. Also it play a risk role in inducing platelet aggregation and promotes arterial thrombosis, increases sympathetic activity and oxidative stress, is an independent risk factor for coronary heart disease and is associated with abnormal fibrinolysis and insulin resistance, all of which are known to promote occlusive cardiovascular events [1]. Moreover, it has been shown that leptin itself induces tissue factor expression in human peripheral blood mononuclear cells and may have a direct role in induction of plasminogen activator inhibitor-1 production, both of which are pivotal agonists of the clotting cascade [1, 3]. Also it has an inflammatory reaction in the vascular wall plays an important role in the development of atherosclerosis and in plaque destabilization and rupture [4].

So that it is very important thing to detect plasma leptin level within those patients who suffer from acute coronary syndrome, in order to detect whether if its elevation can be consider as a main biomarker for future incidence of unstable angina, also to detect the association of leptin level with lipid profile within those patients and what are the main risk factors which play role in elevation of these two levels i.e., leptin level and lipid profile level [1, 2, 5]. So this study tries to detect the main biomarkers for future incidence of unstable angina in Iraqi patients who suffer from coronary syndrome.

Materials and methods:

This is a case control study conducted on (37) unstable angina (UA) patients and 20 healthy control who were closely similar by age, gender and BMI but the only difference that the control were normal non unstable angina subject. Those UA patients were admitted to Baghdad Teaching Hospital and Ibn Al-Bitar Hospital between 2nd September 2010 to 3rd March 2011. Clinical data that related with patients age, gender and BMI were collected from patients' files by specific data sheet form designed for this study. Blood samples were drawn between 7.00-9:00 AM after 10 hours fasting. All specimens were centrifuged at 4000 rpm within 2 hour of collection and serum stored at -20°C until analysis. After preparation of serum, the levels of leptin were measured with (ELIZA) method using DRG (DRG Instruments GmbH\ GERMANY) kit. While the levels of lipid were measured with (Enzymatic) method by using (Biolabo Reagents\FRANCE) Kit. All of the patients who involved in this study were fulfilled the inclusion criteria for this study. The inclusion criteria include

adult UA patient ≥ 18 years old, male or female and were admitted for the first time for the treatment of UA. The exclusion criteria include patients not suffering from diabetes, hypertension, any chronic disease, any current infection, not receiving cortisone and women not in menstrual cycle period.

Statistical analysis:

The statistical tests used were Linear regression test, Correlation test both were used to detect the association and correlation between leptin and lipid profile with several factors and Student t test was used to find if there is a differences between the measured leptin and lipid profile values with the control values. Significant was set at $P < 0.05$. Sampling method used for this study was convenience sampling method. The results were expressed as mean ± SDs.

Results:

Clinical characteristics of unstable angina patients are summarized in (Table-1).

The mean values for age, weight, height, and BMI (body mass index) did not show any difference between the groups of UA and healthy control. As shown in the following table.

Demographic data			
Variables	**UA patients**	**Control**	**P value[*]**
n	37	20	
Gender,male/female	21/16	10/10	
Age (years)	57.2±11.7	55.5±13.1	0.801
Weight (kg)	78.70± 17.42	75.29±14.35	0.572
High (cm)	166.83±12.29	160.46±10.85	0.649
Waist (cm)	120.52±17.56	118.68±13.79	0.472
Lipid Profile **HDL** **LDL** **TG** **TC** **VLDL**	48.04±13.71 138.29±14.37 185.93±11.21 233.85±11.49 31.18±14.91	70.19±11.37 98.22±14.02 92.51±15.73 159.48±13.29 18.50±13.81	0.062 0.014 0.028 0.003 0.001
BMI (km/m^2)	28.31±11.23	27.19±9.24	0.572
Leptin total (ng/ml)	34.25±9.67	27.33±7.20	0.027

Table-1: UA Patients and Healthy Control Variables
*Stusent t test

Depending on the above results in table-1, which show that the age, weight, high, waist and BMI of unstable angina patients and control show no

significant difference since all *P* values were > 0.05. While for lipid profile the results show that Only HDL values do not show significance difference between unstable angina patients and control. While the other factors LDL, TG, TC and VLDL all show significant difference and VLDL show the highest difference. For leptin level the result of student *t* test show significant difference between unstable angina patients and their control.

The risk factors associated with elevation of leptin levels in unstable angina patients is shown in the (Table-2).

Variables of UA Patiens	*P* value	Correlation (*r*)	
Gender	0.226		
Age (years)	0.017	≥ 40	(*P*=0.043) 1.358
		< 40	(*P*=0.794) 0.212
BMI	0.000	Underweight (*p*=0.274) 0.239	
		Normal (*p*=0.407) 0.723	
		Overweight (*p*=0.002) 1.973	
		Obesity (*p*=0.000) 3.116	
Smoking	0.351		

Table-2: Association and Correlation of UA Patients Variables with Leptin Levels
*Linear regression test

According to the above results in (Table-2) which show that gender show insignificant association with leptin i.e., it's not consider as a risk factor for leptin elevation. While for the patient's ages, the results show that the age above or equal to 40 years old is strongly associated with elevation of leptin, but the age group lower than 40 years show insignificant association. So this means that patient age will act as a risk factor only when it is equal or above 40 years old. While in case of BMI according to the results obesity is the highly correlated with elevation of leptin level in UA patients followed by overweight. While normal weight and under weight both show insignificant association with leptin level.

While risk factors associated with lipid profile levels in UA patients is shown in the (Table-3).

Variables of UA Patients	*P* value[*]	Correlation (*r*)

Gender	0.049	Male (*p*=0.027) 2.011 Female (*P*=0.518) 1.901
Age (years)	0.013	≥ 40 (*P*=0.011) 2.298 < 40 (*P*=0.141) 0.336
BMI	0.000	Underweight (*p*=0.113) 0.107 Normal (*p*=0.219) 0.261 Overweight (*p*=0.008) 2.117 Obesity (*p*=0.000) 3.055
Smoking	0.032	Smoking (*p*=0.005) 3.658 Not smoking(*p*=0.621) 0.081

Table-3: Association and Correlation of UA Patients Variables with Lipid Profile Levels
*Linear regression test

According to (Table-3) which show that only male gender has significant association with lipid level within UA patients, patients ages above and equal 40 years old, BMI (overweight and obesity) and smoking show significant association with lipid levels i.e., these variables consider as risk factors which play role within increases of lipid levels. Since their correlations with lipid levels were all in positive correlation.

Association of Lipid with Leptin	*P* value[*]	Correlation (*r*)
Lipid	0.000	HDL (*p*=0.272) 0.467 LDL (*p*=0.029) 2.639 TG (*p*=0.011) 2.414 TC (*p*=0.031) 2.537 VLDL (*p*=0.022)2. 962

Table-4: Association and Correlation of UA Patients Lipid Profile Levels with Leptin Levels
*Linear regression test

The results in (table-4) show that LDL, TG, TC and VLDL show significant association and positive correlation with leptin level i.e., when leptin level will increases these variables will also increases. While HDL show insignificant association with leptin level i.e., leptin level has no effect on HDL level.

Discussion:

The results of student *t* test show significant relationship between lipid profile levels (LDL, TG, TC and VLDL) but not HDL also leptin levels of unstable angina (UA) patients compared with their healthy control i.e., lipid profile levels and leptin level of UA patients higher than that of healthy control.

The elevation of leptin level is considered as a useful biomarker for the incidence of UA in future. Also leptin role is not only limited as an obesity gene product that regulate body weight and energy metabolism, but it has a much boarder biological role. Leptin induces platelet aggregation and promotes arterial thrombosis, increases sympathetic activity and oxidative stress. Thus leptin is considered as an independent risk factor for incidence of UA [1].

This point of detection and consideration for leptin level as a biomarker for future incidence of UA could be considered as a novel point since no such study has been done before on Iraqi UA patients.

The explanation of the significant association and positive correlation between leptin level and lipid profile level is that leptin, the adipose tissue-derived protein product of the obesity gene, is an important multifunctional polypeptide that participates in regulation of body weight and energy metabolism. In human obesity, serum leptin concentrations are directly proportional to body fat mass and lipid profile level mainly triglyceride and total cholesterol[2,5], while TG, TC, LDL and VLDL but not HDL show significant and positive correlation with leptin level. This point has been confirmed by Beltowski and coworkers that HDL has an inverse relationship with leptin level [4]. Moreover the high level of HDL in blood indicates the low chances for incidence of heart diseases and blood vessels problems like UA. While high level of LDL, TG and VLDL in blood indicated high chance of incidence of heart and blood vessels diseases [6].

Therefore according to the results of this study which indicated that increases the level of leptin give an indication of increases harmful lipid particles which are considered as a risk factor for incidence of heart and blood vessels problems in future.

The results show age play role in elevation of leptin level since it show significant association ($P < 0.05$) due to the fact that in aging people there are several factors that start to increase like immunological factors and leptin, age factor plays a role in elevation of leptin level when patients are overweight women, therefore age alone as an isolated factor could not be considered so risky without BMI and gender factors [7].

For the insignificant association between gender and leptin level in our study this mainly is because of the predominant gender is male gender, and the main gender that mainly causes elevation of leptin level is the female gender [8].

Also this study show BMI and specifically obese and overweight is a strong risk factor that play a role in elevation of leptin in UA patients. This point has been confirmed by Mai and his colleague who mentioned that BMI (overweight and obese) is one of the main risk factor in elevation of leptin level[9].

For the insignificant association between smoking and leptin level, this is because the smoking has no effect on leptin level [10].

While for lipid profile the results show that gender and age (specifically old male but not old female) consider as a risk factor in increasing lipid profile level, the main explanation is that cholesterol levels tend to rise in elderly but men tend to develop cardiovascular disease at least 10 to 15 years earlier than women. They also face an increased risk of dying from heart diseases at a much younger age than women [11].

Moreover results show that smoking has significant association and positive correlation with increases of lipid profile level this is because smoking plays a serious role in causing elevation of lipid level [11].

BMI specifically overweight and obesity but not normal or under weight both show significant association and high positive correlation with lipid levels i.e., when the weight increases above normal lipid level will increases too.

This is because people who are overweight do not have enough HDL or (High Density Lipoprotein) but they have high blood cholesterol, triglycerides and LDL. Further, obesity enhances the risk of type 2 diabetes and high blood pressure- which are linked with heart disease i.e., UA[11].

So the main conclusion that can be obtained from this study is that the doctors should focus on leptin level and lipid profile level as biomarkers for future incidence of unstable angina in Iraqi patients.

Acknowledgments:

References:-

1- Amasyali, B.; Kose, S.; Kursaklioglu, H.; Kilic, A. and Sik, E. (2008). Leptin in acute coronary syndromes: Has the time come for its use in risk stratification? .International Journal of Cardiology. 130: 264-265.

2- Al-Daghri, N.; Al-Rubean, K.; Bartlett, W. A.; Al-Attas, O.; Jones, A. F. and Kumar, S. (2003). Serum leptin is elevated in Saudi Arabian patients with metabolic syndrome and coronary artery disease. Diabetic Medicine. 20: 832-837.

3- Dubey, L.; Zeng, H.; Hashmi, S.; Hongjie, W. and Taoa, H. (2008). Association of plasma leptin levels and complexity of the culprit lesion in patients with unstable angina. International Journal of Cardiology. 126: 183-189.

4- Beltowski, J. (2006). Leptin and atherosclerosis. Atherosclerosis .189: 47-60.

5- Wolk, R.; Berger, P.; Lennon, R.J.; Brilakis, E.S. and Johnson, B.D. (2004). Plasma Leptin and Prognosis in Patients with Established Coronary Atherosclerosis. Journal of the American College of Cardiology .44(9): 1819-1824.

6- National Heart Lung and Blood Institute. (2008). High Blood Cholesterol. From: http://www.nhlbi.nih.gov/health/dci/Diseases/Hbc/HBC_WhatIs. html.

7- Mendoza-Nunez, V. M.; Garcı´a-Sa´nchez, A.; Sa´nchez-Rodrı´guez, M.; Galva´n-Duarte, R.E. and Fonseca-Yerena, M. E. (2002). Overweight, Waist Circumference, Age, Gender, and Insulin Resistance as Risk Factors for Hyperleptinemia. Obesity research. 10(4): 235-259.

8- Guler, N.; Kirerleri, E.; Ones, U.; Tamay, Z.; Salmayenli, N. and Darendeliler, F. (2004). Leptin: Does it have any role in childhood asthma? J Allergy Clin Immunol. 114: 254-259.

9- Mai, X. M.; Chen, Y. and Krewski, D. (2009). Does leptin play a role in obesity asthma relationship. Pediatr allergy immunol .20: 207-212.

10- Yoshinari, M.; Wakisaka, M. and Fujishima, M. (1998). Serum Leptin Levels in Smokers Wit h Type 2 Diabetes. Diabetes Care. 21(4): 516-517.

11- Careurheart.com. (2010). Small Lapses on Your Part Can Lead To High Cholesterol Levels! From: http://www.careurheart.com/heart-care-articles/ causes-of-high-cholesterol.htm.