

The impact of inflammation on Resistin, Insulin and Troponin I in Acute Myocardial Infarction patients

Ammal E. Ibrahim *

Hadeef D. EL-Yassin*

Hamid K. AL-Janabi **

BSc, MSc

BSc, PhD (post doctorate)

FRCP, MD, FACC, FCCP

Summary:

Background: Myocardial Infarction (MI) is most commonly due to occlusion (blockage) of a coronary artery following the rupture of a vulnerable atherosclerotic plaque. It has been suggested that the adipose tissue may play an important role in mediating this chronic inflammatory process. Inflammatory responses are involved in the initiation and progression of atherosclerotic plaques. Resistin is a cysteine-rich polypeptide that is expressed at relatively lower levels in human adipocytes but higher levels in macrophages. Insulin is an important hormone as it regulates the level of glucose, in the blood. This protein is formed in specialized cells of the pancreas called beta islet cells.

Subjects and Methods: The study included 50 patients with AMI and forty healthy subjects as controls. Levels of resistin, insulin and troponin were measured.

Results: The levels of resistin and insulin were significantly elevated with ($p < 0.001$), there was a positive correlation between resistin with insulin and troponin in acute myocardial infarction.

Conclusions: There was a significant increase in the levels of resistin, in acute myocardial infarction patients and this increase may be related to inflammation. Elevated levels of resistin can lead to glucose intolerance in AMI patients.

Key word: Resistin, Insulin, Troponin I and Acute Myocardial Infarction.

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Introduction:

Myocardial Infarction (MI) or Acute Myocardial Infarction (AMI), commonly known as a heart attack, is the interruption of blood supply to part of the heart, causing some heart cells to die. This is most commonly due to occlusion (blockage) of a coronary artery following the rupture of a vulnerable atherosclerotic plaque, which is an unstable accumulation of lipids (cholesterol) and white blood cells (especially macrophages) in the wall of an artery. The resulting is ischemia (restriction in blood supply and oxygen shortage) (1). Inflammatory responses are involved in the initiation and progression of atherosclerotic plaques. The majority of inflammatory cells infiltrating the arterial wall in early atherogenesis are monocytes. It has been suggested that adipose tissue may play an important role in mediating this chronic inflammatory process and, subsequently, cardiovascular disease risk and therefore may not only be considered as a storage site for fat, (2). Adipose tissue is increasingly recognized as a key regulator of energy balance, playing an active role in lipid storage and buffering, and synthesizing and secreting a wide range of endocrine products that may be directly involved in the pathogenesis of the complications associated with obesity. So obesity is considered as the major independent risk factor for atherosclerotic cardiovascular disease. Yet the mechanisms that relate fat mass to vascular disease are poorly understood. (2)

Resistin is a cysteine-rich polypeptide that is expressed at relatively lower levels in human adipocytes but higher levels in macrophages. Although animal studies further support this hypothesis, the physiologic relevance of resistin for obesity related conditions in humans remains controversial. (3) Resistin is an adipocytokine that may link insulin resistance, inflammation and atherosclerosis. Resistin up-regulates the expression of adhesion molecules in endothelial cells and promotes smooth muscle cell proliferation. Serum resistin levels were reported to be high in patients with coronary artery disease, especially unstable angina. (4).

Insulin is essential for regulating the energy and glucose metabolism in the body. Insulin causes cells in the liver, muscle, and fat tissue to take up glucose from the blood, storing it as glycogen in the liver and muscle. When insulin is absent, glucose is not taken up by body cells and the body begins to use fat as an energy source. (5) Troponin is attached to the protein tropomyosin and lies within the groove between actin filaments in muscle tissue. In a relaxed muscle, tropomyosin blocks the attachment site for the myosin cross bridge, thus preventing contraction. When the muscle cell is stimulated to contract by an action potential, calcium channels open in the sarcoplasmic reticulum and release calcium into the sarcoplasm. (6)

Subjects and methods:

This study was performed during the period from December 2009 to April 2010. This study included fifty patients with Acute Myocardial Infarction (AMI) admitted to Cardiac Care Unit (CCU) at Medical City

*Dept. of Physiology Chemistry, College of Medicine University of Baghdad.

**Dept. of Internal Medicine, Collage of Medicine, University of Baghdad

Teaching Hospital and Ibn –Al-betar Hospital in Baghdad, with age range (20-78) years. Exclusion criteria included those patients with a history of AMI or diabetes mellitus or any chronic diseases. Control group implied forty age, sex and BMI matched, apparently healthy individuals, were included in this study. From each patient and control, five ml venous blood was aspirated from a suitable vein. Samples were collected between (8-9 A.M.) after 10 hours fasting.

Statistical analysis: Statistical analysis was performed by statisticians with the SPSS 15.01 Statistical Package for Social Sciences and also Excel 2003. Data analysis was done using chi- square test for tables with frequencies, while independent sample t-test was used for tables with means and standard error. *p* value of ≤ 0.05 was used as the level of significance. Correlation coefficient used to find the correlation between studied markers by using Pearson correlation. Descriptive statistics for the clinical and laboratory results were

formulated as mean and standard error. Blood samples were transferred to plain tubes for storage to measure the insulin and resistin. The non heparinized blood in the plain tubes were left to clot and then centrifuged at 4000 rpm for 5 minutes to separate the serum and dispensed into tightly closed Eppendorf tubes in 1.0 ml and stored at -20 C° until assayed. Each serum sample was analyzed, Resistin, insulin and troponin, were measured by using ELISA kits from United States Biological Company.

Results

The results showed that the patients with AMI have significantly higher levels of (resistin and insulin) with (p<0.001) as shown in tables (1)

Table 1: comparison between groups for (resistin, insulin and troponin I)

parameters	Female patients Mean±SR NO.=16	Female Control Mean±SR NO.=16	P-value	Male Patient Mean±SR NO.=34	Male Control Mean±SR NO.=24	p value	Total Patients Mean±SR NO.=50	Total Control Mean±SR NO.40	P- value
Resistin ng/ml	8.18 ±2.19	4.82 ±1.24	<0.001	9.53 ±2.30	4.515 ±1.10	<0.001	9.05 ±2.10	4.64 ±1.93	<0.001
Troponin I ng/ml	34.63 ±6.51	0.0	--	38.18 ±9.95	0.0	--	30.21 ±8.36	0.0	----
Insulin ulU/ml	71.99 ±6.05	29.792 ±8.309	<0.001	71.74 ±9.70	34.092 ±7.980	<0.001	71.83 ±11.09	32.39 ±4.50	<0.001

The results showed that there was strong significant positive correlation between resistin with insulin in female patients group (r= 0.572 with P<0.001), male patients group (r=0.578 with P<0.001) and in total patients (r=0.610 with P<0.001), also between resistin with troponin I in total patients group(r=0.534 with P<0.001) Significant positive correlation was also found between resistin with troponin I in female patients(r=0.480 with P<0.001) and in male patients group (r=0.451 with P<0.001).

Table (2) the correlation between resistin with insulin and troponin I for studied groups.

parameters	Female Patients NO.=16	Female Control NO.=16	Male Patients NO.=34	Male Control NO.=24	Total Patients NO.=50	Total Control NO.=40
Troponin I ng/ml	0.480*	---	0.451*	--	0.534*	----
Insulin ulU/ml	0.572*	0.450	0.578*	0.578	0.610*	0.421

*= (Significantly P with P<0.001)

Discussion:

The results revealed that there was a significantly higher level of resistin among AMI patient in males and females when compared to corresponding sex of control groups there was no significant difference between male patient group with female patients group as shown in table (1) This study agree with study done by Islamabad and Gujranwala in 2010(7) who found strong association between the elevated resistin levels and CAD in a Chinese population. Resistin directly activates the endothelium through up regulation of adhesion molecules, an effect that is antagonized by adiponectin.

Resistin also induces production of the proinflammatory cytokines endothelin-1, monocyte chemoattractant protein (MCP)-1, and pentraxin by endothelial cells. It also promotes migratory activity of vascular smooth muscle cells. In macrophages resistin facilitates lipid accumulation, thereby promoting formation of foam cells; this effect is also antagonized by adiponectin. Furthermore, resistin induces production of the proinflammatory cytokines tumor necrosis factor-α and interleukin (IL)-12 by macrophages through activation of nuclear factor κB. (8). as in table (1) the results

revealed that there was significant increasing in resistin in AMI patients so it agree with this theory. Noboru Toda et al., in 2010(9) mentioned that the attenuated flow responses to sympathetic stimulation likely reflect decreases in NO bioavailability—due to diminished synthesis, accelerated inactivation, or both. Diminished NO bioavailability likely results from a defect in the insulin-signaling pathway downstream to the insulin receptor and includes phosphorylation of insulin receptor substance protein and activation of phosphatidylinositol 3-kinase, leading to phosphorylation of endothelial nitric oxide synthase (eNOS) and (Nitric Oxide) NO production. Low insulin receptor substance levels found in Insulin Resistance likely interfere with normal signal transduction, leading to diminish NO synthesis (9). Elevated level of resistin can lead to insulin resistance because resistin interfere with insulin signal pathway, this was the indirect way of decrease NO production and decrease vasodilatation. Sabir Hussain, et al., in 2010(10) found another effect of resistin on cardiomyocytes, in that resistin could increase the cell surface area of cardiomyocytes and could also be involved in the activation of hypertrophic signal transduction processes (eg, MAPKs). Hyper-resistinemia may contribute to the development of pathologic cardiac hypertrophy; resistin-transduced ventricular cardiomyocytes lead to the impairment of myocytes relaxation. A negative effect of resistin on the cardiac function is also supported by the association of the cytokine with high risk in patients with congestive heart failure. (10), from the results we noticed that the patients with higher level of resistin have the longest time to stay in CCU. So according to the relation of resistin with NO that mentioned by Noboru Toda et al., 2010,(9)

Resistin with insulin: As shown in table (1) it was found that insulin is significantly higher in patients group than control groups. To our knowledge this is the first study reported insulin level in AMI patients. In this study it was found that there was significantly positive correlation between resistin with insulin as shown in table (2). Insulin forces arterial wall muscle to relax, increasing blood flow, especially in micro arteries; lack of insulin reduces flow by allowing these muscles to contract.(11) Circulating insulin levels also have important effects on myocardial flow at rest and during pharmacologically stimulated myocardial hyperemia. Infusion of insulin in healthy subjects and in patients with diabetes raises myocardial flow at rest. Importantly, acute insulin infusion also evokes significant flow increases in patients with coronary artery disease (12) more tenuously; hyperinsulinaemia might be responsible for elevating blood pressure through actions on renal sodium retention or the sympathetic nervous system. Yet there appear few mechanisms whereby hyperinsulinaemia or insulin resistance might increase glomerular loss of albumin, or endothelial cell or acute phase activation. (13) Mechanisms accounting for the progressively worse coronary circulatory functional abnormalities observed in more severe states of insulin resistance (IR) remain unclear. They may be related to an increased number of

vascular stressors, including elevated plasma levels of free fatty acids, triglycerides, oxidized low-density lipoprotein cholesterol, and oxidation-prone small dense low-density lipoproteins. Pro-inflammatory cytokines and inflammatory markers such as C-reactive protein and adipocyte-derived adipokines contribute to further decreases in production and inactivation of NO. Formation of reactive oxygen species in hyperglycemia, together with inactivation of protein kinase C, may further impair vascular function. (14) Endothelial dysfunction, even in the absence of macrovascular coronary artery lesions, may be responsible for reduction in or failure to appropriately augment coronary flow, leading to myocardial ischemia. When coexisting with obstructive coronary artery disease, endothelial dysfunction may cause more severe and extensive stress-induced perfusion abnormalities, possibly because of stress-related sympathetically mediated vasoconstriction. (13), that agree with what had been noticed in this study that the patients with elevated level of insulin have high blood pressure. The vasodilation evoked by insulin is mediated by NO released from the endothelium by eNOS. It is well known that insulin can induce Akt phosphorylation through the activation of the IRS-1/phosphatidylinositol 3-kinase (PI3K) cascade that occurs after the physical interaction between IRS-1 and the PI3K subunit p85. Insulin able to stimulate interaction between IRS-1 and p85. (15) Endothelial exposure to resistin significantly decreased IRS-1 intracellular levels and markedly impaired the ability of insulin to induce IRS-1/PI3K interaction by blunt the insulin signaling pathway acting on both IRS-1 levels and its ability to activate PI3K also Resistin can increase endothelial IRS-1 ubiquitination. (16) In vitro studies in human adipose cells supports the role of resistin in reducing glucose uptake, revealing a potential functional role for resistin in vivo metabolism. (15) And these suggest that resistin may have a dual role involved in sub-clinical inflammation as well as in altering glucose metabolism leading to the progression of T2DM. Resistin suppresses insulin-stimulated glucose uptake in cultured adipocytes, and this effect is prevented by exposure to anti-resistin antibodies. Finally, treatment with these antibodies decreases blood glucose and improves insulin sensitivity in obese mice. All these data suggest that resistin could contribute to the insulin resistance observed in obese subjects by decreasing insulin sensitivity at least in skeletal muscle tissue. (15) Conversely, transgenic over expression of resistin in mice increases hepatic glucose production without decreasing skeletal muscle glucose utilization. Taken together, these data suggest that the liver is a major target of resistin actions (Christophe Graveleau et.al. 2009) (19) have shown that isolated cardiomyocytes are insulin-responsive and share many characteristics of adipocytes and skeletal muscle in terms of insulin stimulation of glucose transport. (Shamina M. Rangwala et al 2009)(20) in vivo experiments have suggested that resistin impairs glucose tolerance by inhibiting the ability of insulin to suppress hepatic glucose production as well as by inhibiting glucose uptake in skeletal

muscle. (20) The results from (Christophe et al. 2009)(19) study indicate that resistin also alters glucose uptake in cardiac muscle (Christophe et al. 2009). (19) Observed that human resistin caused a significant reduction in insulin stimulated glucose uptake in cardiomyocytes. Insulin increases myocardial glucose utilization and reduces free fatty acid (FFA) oxidation. Glucose oxidation is more oxygen efficient than FFA oxidation, hence this change in myocardial metabolism is theoretically beneficial to the heart especially during ischemia, and is thought to be the main mechanism behind the beneficial effects seen in the various trials of glucose-insulin-potassium infusions. Also, insulin resistance has been shown to have several adverse effects on the metabolism of the myocardium that heighten the effects of ischemia. These include reduced glucose uptake and oxidation, increased FFA uptake and oxidation as well as decrease in calcium transport within the sarcolemma and alterations in myofibrillary regulatory contractile proteins. The net effect is a reduction in cardiac efficiency that causes increases the susceptibility of the insulin resistant heart to myocardial ischemia and to a greater reduction in myocardial performance (21) Therefore, if human resistin impairs insulin action in insulin-responsive cells in vitro, then increased concentrations of resistin in humans could potentially impair insulin sensitivity in vivo and contribute to the association between hyper-resistinemia and glucose intolerance. (21) As shown in table (2) there was significantly positive correlation between resistin with troponin. Troponin in this study is used to measured the infarction size so as resistin is positively correlated with troponin that mean the patients with higher levels of resistin will have higher level from troponin and bigger infarction size so this patient will have bad or slow progress to cure. Troponin is a much more sensitive measure of cardiac damage than the conventional MB isoenzyme fraction of myocardial creatine kinase. Cardiac troponin levels in the blood predict short-term prognosis in acute coronary syndromes particularly in microinfarction. Troponin I has also been shown to inhibit angiogenesis in vivo and in vitro. (22)

Conclusions:

Elevated levels of resistin can lead to glucose intolerance in AMI patients because it interfere with signal transduction of insulin.

References:

1. Kosuge, M; Kimura K, Ishikawa T et al. "Differences between men and women in terms of clinical features of ST-segment elevation acute myocardial infarction". *Circulation Journal*. March 2006; 70 (3): 222-226.
2. Abdulwahab N. Al-Isa, Lukman Thalib, Abayomi O. Akanji *Circulating markers of inflammation and endothelial dysfunction in Arab adolescent subjects: Reference ranges and associations with age, gender, body mass and insulin sensitivity".* *Atherosclerosis* ;2010;208(2): 543-549)

3. Christin Heidemann, Rob M. van Dam, James B. Meigs." *Total and High-Molecular-Weight Adiponectin and Resistin in Relation to the Risk for Type 2 Diabetes in Women.*" *Ann Intern Med.*;2008;149:307-316.
4. Yukihiko Momiyama, Reiko Ohmori, Ryuichi Kato, Hiroaki Taniguchi, Haruo Nakamura, Fumitaka Ohsuzu." *Prognostic significance of serum resistin levels in patients undergoing percutaneous coronary intervention.*" *JACC*: 2010;55 100- 126.
5. De la Monte SM, Wands JR. "Review of insulin and insulin-like growth factor expression, signaling, and malfunction in the central nervous system: relevance to Alzheimer's disease". *J. Alzheimers Dis. February 2005; 7 (1): 45-61.*
6. Ammann P, Pfisterer M, Fehr T, Rickli H. "Raised cardiac troponins." *BMJ*. 2004;328:1028-9. PMID 15117768.
7. Islamabad and Gujranwala." *Resistin gene promoter region polymorphism and the risk of hypertrophic cardiomyopathy in patients". (Translational Research 2010;155:142-147.*
8. Giamila Fantuzzi and Theodore Mazzone." *Adipose Tissue and Atherosclerosis."Arterioscler Thromb Vasc Biol.* 2009;27:996-1003.
9. Noboru Toda, Takeshi Imamura, Tomio Okamura." *Alteration of nitric oxide-mediated blood flow regulation in diabetes mellitus". Pharmacology&Therapeutics;2010;127(3):189-209*
10. Sabir Hussain, Muhammad Asghar, Qamar Javed." *Resistin gene promoter region polymorphism and the risk of hypertrophic cardiomyopathy in patients." Translational Research; 2010:155(3):142-147)*
11. M. Žourek, P. Kyselová, J. Mudra, M. Krčma, Z. Jankove, S. Lacigová, J. víšek, Z. Rušavý." *The Relationship between Glycemia, Insulin and Oxidative Stress in Hereditary Hypertriglyceridemic Rat". Korean Circ J :2009;39:180-184*
12. Lautamäki R, Airaksinen KE, Seppänen M, et al." *Insulin improves myocardial blood flow in patients with type 2 diabetes and coronary artery disease." Diabetes :2009;55(5)11- 26.*
13. Nicolas Wiernsperger, Alain Geloën, Jean-Robert Rapin ." *fructose and cardiometabolic disorders: the controversy will, and must, continue." Clinics. 2010;65(7):729-38.*
14. Heinrich R. Schelbert ." *Coronary Circulatory Function Abnormalities in Insulin Resistance Insights From Positron Emission Tomography."Journal of the American College of Cardiology 2009;53:3-8*
15. N Al-Daghri, R Chetty, PG McTernan, K Al-Rubean, O Al-Attas, AF Jones and S Kumar." *Serum resistin is associated with C-reactive protein and LDLcholesterol in type 2 diabetes and coronary artery disease in a Saudi population." Cardiovascular Diabetology: 2009;4(10):126-135*
16. Maria Teresa Gentile, Carmine Vecchione, Gennaro Marino, Alessandra Aretini, Alba Di Pardo, Giovanna Antenucci, Angelo Maffei, Giuseppe Cifelli, Luca Iorio, Alessandro Landolfi, Giacomo Frati, and Giuseppe Lembo." *Resistin Impairs Insulin-Evoked Vasodilation." Diabetes: March 2008; 57:577-583.*

17. Goran Koraćević, Sladjana Petrović, Miloje Tomašević, Svetlana Apostolović, Miodrag Damjanović. "Stress hyperglycemia in acute myocardial infarction." *Medicine and Biology*; 2010: 13,(3); 152 - 157

18. Odile Gabay and Francis Berenbaum. "Adipokines in Arthritis: New Kids on the Block." *Current Rheumatology Reviews*, 2009;5(4) ;:229-230

19. Christophe Graveleau, Vlad G. Zaha, Arash Mohajer, Ronadip R. Banerjee, Nicole Dudley-Rucker, Claire M. Steppan, Michael W. Rajala, Philipp E. Scherer, Rexford S. Ahima, Mitchell A. Lazar, and E. Dale Abel. "Mouse and Human Resistins Impair Glucose Transport in Primary Mouse Cardiomyocytes, and Oligomerization Is Required for This Biological Action." *Journal of biological chemistry.*;2009: 280(36); 31679–31685.

20. Shamina M. Rangwala, A. Sophie Rich, Ben Rhoades, Jennifer S. Shapiro, Silvana Obici, Luciano Rossetti and Mitchell A. Lazar. "Abnormal Glucose Homeostasis due to Chronic Hyperresistinemia." *Diabetes August 2009*; 53 (8) 1937-1941

21. Patrick M Heck, Stephen P Hoole, Sadia N Khan and David P Dutka Heck et al. "Hyperinsulinemia improves ischemic LV function in insulin resistant subjects" *Cardiovascular Diabetology* :2010;9:27-34

22. P. R. J. Ames, I. Antinolfi, A. Ciampa, J. Batuca, G. Scenna, L. R. Lopez, J. Delgado Alves L. Iannaccone and E. Matsuura. "Primary antiphospholipid syndrome: a low-grade auto-inflammatory disease?". *Rheumatology* 2008;47:1832–1837