

## Underlying Autoimmune Activation Rendering Paradoxical Non-Linear Relationship between Biomarkers and Cardiometabolic Risk

Altan Onat<sup>1\*</sup>, Adnan Kaya<sup>2</sup>, Barış Şimşek<sup>3</sup>, Tuğba Akbaş-Şimşek<sup>4</sup> and Hüsnüye Yüksel<sup>1</sup>

<sup>1</sup>Department of Cardiology, Cerrahpaşa Medical Faculty, Istanbul University, Turkey

<sup>2</sup>Suruç State Hospital, Suruç, Şanlıurfa, Turkey

<sup>3</sup>Section of Cardiology, Siyami Ersek Center for Cardiovascular Surgery, Istanbul, Turkey

<sup>4</sup>Bağcılar Educational Hospital, Istanbul, Turkey

\*Corresponding author: Altan Onat, Professor, Departments of Cardiology, Cerrahpaşa Medical Faculty, Istanbul University, Nispetiye cad. 59/24, Etiler 34335, Istanbul, Turkey, Tel: 90 212 351 6217; Fax: 90 212 351 4235; E-mail: [alt\\_onat@yahoo.com.tr](mailto:alt_onat@yahoo.com.tr)

Received date: January 13, 2016; Accepted date: February 08, 2016; Published date: February 11, 2016

Copyright: © 2016 Onat A, et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

### Abbreviations

AF: Atrial Fibrillation; LDL: Low-Density Lipoprotein; apo: Apolipoprotein; Lp(a): Lipoprotein(a); CHD: Coronary Heart Disease; LpPLA<sub>2</sub>: Lipoprotein-Associated Phospholipase A<sub>2</sub>; CVD: Cardiovascular Disease; MetS: Metabolic Syndrome; HbA1c: Glycated Hemoglobin A1c; MIF: Migration Inhibitory Factor; HDL: High-Density Lipoprotein

Several chronic diseases share in our opinion a common pathophysiologic mechanism (what we designate as autoimmune activation), especially though not exclusively in elderly people prone to impaired glucose tolerance or metabolic syndrome (MetS). This process renders non-linear relationships between a biomarker and cardiometabolic risk.

Numerous observations have been reported which puzzled the observers in offering plausible explanation. These *a priori* paradoxical phenomena relate to circulating proteins (or polypeptides) and include lipoprotein a (Lp (a)), apolipoprotein B (apoB), lipoprotein-associated phospholipase A<sub>2</sub> (Lp-PLA<sub>2</sub>, or PAF-AH), glycated hemoglobin A1c (HbA1c), creatinine, uric acid, thyroid stimulating hormone, acylation stimulating protein, asymmetric dimethyl arginine, macrophage migration inhibitory factor (MIF) and, possibly other serum proteins such as fibrinogen or apoCIII in high-density lipoproteins. We define autoimmune activation as a potentially reversible chronic (serologic) process developing under persistent pro-inflammatory state/oxidative stress rendering mild damage to one or more polypeptides or proteins which are thence perceived as foreign bodies, followed by aggregation to them of protective serum proteins.

### Individual plasma proteins epidemiologically identified as having sustained oxidative damage

In the large prospective Women's Health Study [1], Lp(a) was found inversely associated to the development of type-2 diabetes, with roughly 25% higher relative risk in the bottom quintile compared with the remainder of the sample. This observation, confirmed in the Norfolk-EPIC study [2], can be explained by a mechanism of immune complex formation involving Lp(a) and interfered assay results due to failure by capture antibodies.

Elevated levels of apoB are a well-known risk factor for atherosclerosis complications but low levels of low-density lipoprotein (LDL)-cholesterol [3,4] have been reported to independently predict atrial fibrillation (AF). The large Women's Health Study [3] published

analysis on predictive biomarkers for 795 of incident AF events. AF was predicted inversely by LDL-cholesterol yielding a 28% lower risk of AF per 1-SD increment in LDL-cholesterol, or *vice versa*. The number of small LDL particles was the driver of the stated association. This common arrhythmia is predicted by low apoB levels in an as yet unpublished work in the Turkish Adult Risk Factor study.

Lp-PLA<sub>2</sub>, or PAF-AH, an enzyme concentrated on plasma lipoprotein(a), a major instigator of enhanced inflammation and autoimmune activation, is sensitive to enhanced pro-inflammatory state. The enzyme epitope may sustain damage, inducing aggregation and autoimmune complex formation with a protective protein. It partly escapes immunoassay, resulting in apparently "reduced" mass (and/or activity), and further fails to exhibit anticipated relationship to inflammation markers or cardiometabolic risk [5].

In a cross-sectional analysis of 825 patients with cardiovascular disease, Mayer Jr. and associates [6] observed inverse relationships of Lp-PLA<sub>2</sub> with high fasting glycemia, HbA1c and with quartiles of LDL-C/apoB ratio. Authors concluded that presence of diabetes was independently associated with lower likelihood of elevated Lp-PLA<sub>2</sub>. Previously, in a cross-sectional analysis of about 1000 elderly Swedish subjects, Lp-PLA<sub>2</sub> activity was found not to be independently related to carotid atherosclerosis and to the amount of arterial stenosis [7]. Authors considered that Lp-PLA<sub>2</sub> levels may not constitute a good indicator of the activity within atherosclerotic lesions. We had documented in a population-based sample that circulating Lp-PLA<sub>2</sub> mass was independently associated with prevalent and incident coronary heart disease (CHD) in men, though being inversely associated only with diabetes in men, and only with MetS in women [8].

High-density lipoprotein (HDL)-apoCIII in 120 CHD patients using statins was significantly higher than in 80 control patients (not using) and was an independent predictor of CHD likelihood [9]. HDL-apoCIII significantly rose further in CHD patients after statin therapy. HDL-apoCIII correlated significantly with plasma triglycerides in both groups, but correlation in CHD patients was abolished after statin treatment. Total and LDL-cholesterol tended to be lower in the CHD group and LDL-cholesterol tended to be inversely associated with CHD, while apoB levels were significantly and positively associated with CHD likelihood. These findings suggest that HDL-apoCIII may assume atherogenic properties (just as it was shown in 2008 by us [10] to assume diabetogenic properties) in an environment of low LDL-cholesterol (and high apoB), consistent with autoimmune involvement of Lp(a) protein, but not of apoB. Moreover, of interest is the finding of

further elevation of HDL-apoCIII upon statin therapy (which might have damaged apo(a) of Lp(a)).

Hemoglobin A1c (HbA<sub>1c</sub>) is a measure of glucose control in diabetic persons and a test recommended for diagnosing diabetes at a threshold  $\geq 6.5\%$  [11]. Though several studies demonstrated a linear relationship between HbA<sub>1c</sub> and several outcomes throughout the range of values [12,13], numerous recent studies showed that low HbA<sub>1c</sub> in nondiabetic [14,15] or diabetic individuals [16,17] is associated with increased mortality. Such association suggests the involvement of HbA<sub>1c</sub> levels in autoimmune activation and may be confined to one gender (e.g. to men among Turks). During a follow-up of 9.4 years in ~29,000, both low and high levels of HbA<sub>1c</sub> were associated with a higher risk of cardiovascular disease (CVD) in a Japanese non-diabetic general population [18]. Compared with HbA<sub>1c</sub> levels of 5.0 to 5.4%, HR for CVD in participants without known diabetes were 1.50 (95% CI 1.15-1.95) for HbA<sub>1c</sub> levels of  $<5.0\%$ . This nonlinear relation persisted after excluding people with kidney dysfunction, liver dysfunction, anemia, body mass index  $<18.5$  kg/m<sup>2</sup>, or early events.

Low serum creatinine levels ( $<0.7$ / $<0.8$  mg/dl) may represent a clue to the existence of autoimmune activation [19,20]. This may again manifest itself in one sex in the general population. Increasing serum creatinine values were strongly associated with CHD risk in men but not in women in whom the risk curve was U-shaped [19]. In the prediction of incident type-2 diabetes risk by baseline estimated glomerular filtration rate in the Insulin Resistance Atherosclerosis Study, other than the bottom quintile, the top estimated glomerular filtration rate quintile disclosed excess multi-adjusted risk compared with the referent 4th quintile [21]. The phenomenon of low creatinine levels underlies some hitherto unexplained observations. Renal “hyperfiltrators” represent individuals with autoimmune activation (involving serum creatinine partly escaping assay) and are misclassified into a category of “optimal” renal function. By contrast, they are at significantly higher risk of cardiopulmonary events and death due to the underlying autoimmune process [20].

Chronic inflammatory processes can induce a chronic increase of interleukin-6 and thereby turn the acute phase reaction into a chronic perpetuating state with increased levels of fibrinogen [22]. A large prospective study on urban Chinese (n=6209) demonstrated that MetS was independently predicted in the female but not male [23]. Previously, it had been shown among Turks that independent prediction of MetS by plasma fibrinogen was confined to men [24]. These findings are consistent with the notion that ethnicity-specific sex-dependent prominent autoimmune activation may abolish the linear association of fibrinogen and render manifestation of MetS only in one sex. This forms an example also to prominent autoimmune activation predisposing the opposite sex in different races.

A large prospective study in Taiwan reported that sex- and age-adjusted hazard ratios of uric acid levels for all-cause and cardiovascular mortality were increased not only for elevated uricemia, but also for levels lower than 5 mg/dl [25]. An as yet unpublished work among Turks from applicants of a tertiary hospital showed inverse association of serum uric acid with adverse outcomes, consistent with involvement of uric acid mass in autoimmune activation.

Other serum proteins have also been reported to illustrate instances of autoimmune activation: these include thyroid stimulating hormone

[26], acylation stimulating protein (as yet unpublished), asymmetric dimethyl arginine [27], cystatin-C [28], macrophage MIF [29].

### Enhanced pro-inflammatory state and dysfunction of protective proteins

An important component of the autoimmune process with adverse outcomes is enhanced pro-inflammatory state to which gradual loss of protective properties of certain plasma proteins significantly contribute. ApoA-I, apoA-II, apoE, adiponectin, sex hormone-binding globulin constitute these protective plasma proteins [5]. As an illustration of this point may be given that, in a study on type-2 diabetes incidence in Finnish men, it was confirmed that apoA-I not only failed to significantly protect, but that apoA-I/HDL-C ratio was highly predictive of this outcome [30] suggesting that immature buoyant apoA-I had little insulin-sensitizing properties.

The presumed explanation to the non-linear relationship of a biomarker leading to “paradoxically” elevated cardiometabolic risk is that, plasma proteins sustaining epitope damage due to oxidative stress, partly escape immunoassay, are assayed lower than the actual mass, but have the capacity to render risk of cardiometabolic disease and death.

Autoimmune activation presumably mediates the outcome primarily *via* enhanced low-grade systemic inflammation and endothelial dysfunction. The link between the individual proteins involved in the autoimmune process and the specific cardiometabolic disease remains to be elucidated. Nonetheless, the huge public health issue requires better delineation in the future of damaged plasma proteins by new immunoassay methods or particle/mass measurement by MR spectroscopy.

We conclude that the practicing physician should best start to look for such clues in his/her patients, especially in elderly ones, with the ultimate aim of obtaining deeper insight into their longstanding health problems.

### Acknowledgement

The financial support to the Turkish Adult Risk Factor study by Turkish Automotive Company TOFAŞ, İstanbul is appreciated.

### References

1. Mora S, Kamstrup PR, Rifai N, Nordestgaard BG, Buring JE, et al. (2010) Lipoprotein(a) and risk of type 2 diabetes. *Clin Chem* 56: 1252-1260.
2. Ye Z, Haycock PC, Gurdasani D, Pomilla C, Boekholdt SM, et al. (2014) The association between circulating lipoprotein(a) and type 2 diabetes: is it causal? *Diabetes* 63: 332-342.
3. Lopez FL, Agarwal SK, Maclellan RF, Soliman EZ, Sharrett AR, et al. (2012) Blood lipid levels, lipid-lowering medications, and the incidence of atrial fibrillation: the atherosclerosis risk in communities study. *Circ Arrhythm Electrophysiol* 5: 155-162.
4. Mora S, Akinkuolie AO, Sandhu RK, Conen D, Albert CM (2014) Paradoxical association of lipoprotein measures with incident atrial fibrillation. *Circ Arrhythm Electrophysiol* 7: 612-619.
5. Onat A, Can G (2014) Enhanced proinflammatory state and autoimmune activation: a breakthrough to understanding chronic diseases. *Curr Pharm Des* 20: 575-584.
6. Mayer O Jr, Seidlerová J, Filipovský J, Timoracká K, Bruthans J, et al. (2014) Unexpected inverse relationship between impaired glucose metabolism and lipoprotein-associated phospholipase A2 activity in patients with stable vascular disease. *Eur J Intern Med* 25: 556-560.

7. Lind L, Simon T, Johansson L, Kotti S, Hansen T, et al. (2012) Circulating levels of secretory- and lipoprotein-associated phospholipase A2 activities: relation to atherosclerotic plaques and future all-cause mortality. *Eur Heart J* 33: 2946-2954.
8. Onat A, Hergenç G, Can G, Uğur M, Nartop F (2011) Dual activity of serum lipoprotein-associated phospholipase A(2) yielding positive and inverse associations with cardiometabolic risk. *Clin Chem Lab Med* 49: 1349-1357.
9. Xiong X, Liu H, Hua L, Zhao H, Wang D, et al. (2015) The association of HDL-apoCIII with coronary heart disease and the effect of statin treatment on it. *Lipids Health Dis* 14: 127.
10. Onat A, Hergenç G, Ayhan E, Uğur M, Kaya H, et al. (2009) Serum apolipoprotein C-III in high-density lipoprotein: a key diabetogenic risk factor in Turks. *Diabet Med* 26: 981-988.
11. American Diabetes Association (2010) Diagnosis and classification of diabetes mellitus. *Diabetes Care* 35(Suppl 1): S62-S69.
12. Khaw KT, Wareham N, Luben R, Bingham S, Oakes S, et al. (2011) Glycated hemoglobin, diabetes, and mortality in men in Norfolk cohort of European Prospective Investigation of Cancer and Nutrition (EPIC-Norfolk). *BMJ* 322: 15-18.
13. Schöttker B, Müller H, Rothenbacher D, Brenner H (2013) Fasting plasma glucose and HbA1c in cardiovascular risk prediction: a sex-specific comparison in individuals without diabetes mellitus. *Diabetologia* 56: 92-100.
14. Carson AP, Fox CS, McGuire DK, Levitan EB, Laclaustra M, et al. (2010) Low hemoglobin A1c and risk of all-cause mortality among US adults without diabetes. *Circ Cardiovasc Qual Outcomes* 3: 661-667.
15. Aggarwal V, Schneider AL, Selvin E (2012) Low hemoglobin A(1c) in nondiabetic adults: an elevated risk state? *Diabetes Care* 35: 2055-2060.
16. Altay S, Onat A, Karadeniz Y, Özpamuk-Karadeniz F, Can G (2015) Prediction by Low Plasma HbA1c of Mortality, Cardiac and Noncardiac Disease Risk: Modulation by Diabetic Status and Sex. *J Investig Med* 63: 821-827.
17. Colayco DC, Niu F, McCombs JS, Cheetham TC (2011) A1C and cardiovascular outcomes in type 2 diabetes: a nested case-control study. *Diabetes Care* 34: 77-83.
18. Goto A, Noda M, Matsushita Y, Goto M, Kato M, et al. (2015) Hemoglobin a1c levels and the risk of cardiovascular disease in people without known diabetes: a population-based cohort study in Japan. *Medicine (Baltimore)* 94: e785.
19. Onat A, Can G, Ademoğlu E, Çelik E, Karagöz A, et al. (2013) Coronary disease risk curve of serum creatinine is linear in Turkish men, U-shaped in women. *J Investig Med* 61: 27-33.
20. Altay S, Onat A, Özpamuk-Karadeniz F, Karadeniz Y, Kemalöglu-Öz T, et al. (2014) Renal "hyperfiltrators" are at elevated risk of death and chronic diseases. *BMC Nephrol* 15: 160.
21. Lorenzo C, Nath SD, Hanley AJ, Abboud HE, Gelfond JA, et al. (2009) Risk of type 2 diabetes among individuals with high and low glomerular filtration rates. *Diabetologia* 52: 1290-1297.
22. Gabay C, Kushner I (1999) Acute-phase proteins and other systemic responses to inflammation. *N Engl J Med* 340: 448-454.
23. Ding L, Zhang C2, Zhang G2, Zhang T, Zhao M, et al. (2015) A new insight into the role of plasma fibrinogen in the development of metabolic syndrome from a prospective cohort study in urban Han Chinese population. *Diabetol Metab Syndr* 7: 110.
24. Onat A, Ozhan H, Erbilen E, Albayrak S, Küçükdurmaz Z, et al. (2009) Independent prediction of metabolic syndrome by plasma fibrinogen in men, and predictors of elevated levels. *Int J Cardiol* 135: 211-217.
25. Kuo CF, See LC, Yu KH, Chou IJ, Chiou MJ, et al. (2013) Significance of serum uric acid levels on the risk of all-cause and cardiovascular mortality. *Rheumatology (Oxford)* 52: 127-134.
26. Onat A, Aydin M, Can G, Çelik E, Altay S, et al. (2015) Normal thyroid-stimulating hormone levels, autoimmune activation, and coronary heart disease risk. *Endocrine* 48: 218-226.
27. Onat A, Köroğlu B, Can G, Karagöz A, Yüksel M, et al. (2014) Apparently "low" serum asymmetric dimethylarginine is associated with fasting glucose and tends toward association with type-2 diabetes. *Anadolu Kardiyol Derg* 14: 26-33.
28. Shi GP, Sukhova GK, Grubb A, Ducharme A, Rhode LH, et al. (1999) Cystatin C deficiency in human atherosclerosis and aortic aneurysms. *J Clin Invest* 104: 1191-1197.
29. Çoban N, Onat A, Yildirim Ö, Can G, Erginel-Ünaltuna N (2015) Oxidative stress-mediated (sex-specific) loss of protection against type-2 diabetes by macrophage migration inhibitory factor (MIF) -173G/C polymorphism. *Clin Chim Acta* 438: 1-6.
30. Fizelova M, Miilunpohja M, Kangas AJ, Soinen P, Kuusisto J, et al. (2015) Associations of multiple lipoprotein and apolipoprotein measures with worsening of glycemia and incident type 2 diabetes in 6607 non-diabetic Finnish men. *Atherosclerosis* 240: 272-277.