Original article **UDC: 616.379-008.64-053.2:616.61-008.6]:616-008.9:577**
doi:10.5633/amm.2017.0315

THE IMPACT OF MECHANISMS OF OXIDATIVE STRESS ON THE DEVELPOMENT OF DIABETIC NEPHROPATHY IN TYPE 1 DIABETES

Jelena Vučić[1], Sandra Stanković[1], Karin Vasić[1], Tatjana Cvetković[2,3]

In a series of 50 type 1 diabetes participants, mean age 18.9±2.8 years, disease duration longer than five years, each with proven incipient diabetic nephropathy, our goal was to determine the potential impact of oxidative stress on the development of diabetic nephropathy. We determined the antioxidant activity of thiol compounds (SH groups), lipid peroxidation by measuring malondialdehyde (MDA), and advanced oxidation protein products (AOPP). Insufficient antioxidant defense was proven in the diabetic nephropathy group, with a dramatic drop in the activity of thiol compounds compared to controls (132.32±36.60 μmol/L vs 189.22±42.90 μmol/l, p <0.001). This may explain the increase in oxidative stress, or increased lipid peroxidation characterized by the production MDA in the diabetic nephropathy group compared to controls (51.28±12.76 μmol/L vs 17.54±6.35 μmol/L, p <0.001), and the increase in AOPP in patients compared to controls (48.82±13.84 vs 18:45±1.73 μmol/l, p <0.001). Correlation analysis showed a correlation between MDA and SH (r=-0.451, p <0.001), and between SH and AOPP (r=-0.487, p <0.001). However, only MDA is a statistically significant risk factor for the development of diabetic nephropathy. Univariate logistic regression analysis showed that MDA is an independent risk factor. An increase in MDA by 1μmol increases the risk of the development of diabetic nephropathy by 32.4%. Reducing SH concentration by 1μmol/L increases the risk of developing diabetic nephropathy by 4%. We conclude that the lower the antioxidant effect of thiol compounds, the more intensive the lipid peroxidation (MDA) will be, thereby increasing the risk for the development of diabetic nephropathy. *Acta Medica Medianae 2017;56(3):94-100.*

Key words: *diabetic nephropathy, oxidative stress, thiol compounds (SH), malondialdehyde (MDA), advanced oxidative protein products (AOPP)*

Clinic of Children's Internal Diseases, Clinical Center Niš, Niš, Serbia[1]
University of Niš, Faculty of Medicine, Institute of Biochemistry, Niš, Serbia[2]
Clinic of Nephrology, Clinical Center Niš, Niš, Serbia [3]

Contact: Jelena Vučić, Studenička 34, Niš, Serbia
jvucic70@gmail.com

Introduction

Type 1 diabetes is undoubtedly a representative of pathological conditions associated with increased oxidative stress. The reactive oxygen species (ROS) can decisively affect not only the occurrence of disease but also the emergence of chronic diabetic complications (1).

Mechanisms of destruction of β-insular cells by ROS are many (1). Under the influence of ROS, peroxidation of polyunsaturated fatty acids occurs with the production of lipid radicals, which in combination with molecular oxygen form lipid peroxide and lipid hydroperoxide radicals. Lipid peroxidation represents the most pronounced negative phenomenon in the functioning of ROS (2). This process is initiated, in most instances, by a hydroxy radical, less often by hydrogen peroxide. It is characterized by peroxidation of polyunsaturated fatty acids with consecutive decomposition of fatty acids and biological membranes leading to cell degradation and cell death. Highly reactive secondary products of lipid peroxidation react with free SH and NH_2 groups of amino acids, peptides, proteins, and nucleotides, thereby modifying the functions of these macromolecules. Advanced oxidation protein products (AOPP) are reliable markers of oxidative stress since all plasma proteins represent a potential target for the action of oxidative radicals. However, numerous mechanisms of antioxidant defense exist (3).

Oxidative modification of proteins, carbohydrates, DNA and lipids is a biological process,

however, when intensified, it leads to cell injury, particularly at the level of the cell membrane. On the other hand, the role of free radicals in physiological processes makes them a mandatory prerequisite of life. Thus, during evolution, a mechanism for defense - the antioxidant system was established. From a functional point of view, antioxidant defense functions at three levels. The first level of antioxidant defense includes systems which to a great extent inhibit endogenous production of free radicals. Proteins such as transferin, oxyhemoglobin, lactoferrin, ferritin and hemosiderin, possess such antioxidant effects. By binding iron, these proteins prevent its entry into the Fenton reaction, a similar effect is manifested by ceruloplasmin, which binds copper. The second level of defense is active in the normal state and increases the production of reactive oxygen species. This level can be divided into enzymatic and non-enzymatic pathways (4). Enzymes (superoxide dismutase, catalase, glutathione peroxidase, glutathione reductase, glutathione S-transferase) make up, the so-called, first line of antioxidant protection, while non-enzymatic antioxidants (glutathione, vitamins C and E, beta carotene, thiol compounds - glutathione, methionine and cysteine, albumin, taurine and its precursors, bilirubin, uric acid, estrogens, creatine, coenzyme Q, polyamines, flavonoids and other phenolic compounds represent a secondary line of defense. Thiol compounds such as glutathione, cysteine and homocysteine serve as biological reduction systems of the cell. The term "thiol" refers to the SH group, which determines the chemical characteristics of these compounds, enabling them to be involved in very important metabolic pathways. Glutathione oxidizes with lipid peroxides giving glutathione peroxide; this reaction is catalyzed by glutathione peroxidase. This demonstrates that glutathione can neutralize lipid peroxides. Glutathione cellular concentration, including concentration in vascular cells, is crucial in regulating its antioxidant role (4).

The occurrence of diabetes mellitus, as well as the development of complications, is closely related to an imbalance in pro/antioxidant status of the cell which disrupts the cell's redox potential. Oxidative stress in diabetes mellitus is caused by excessive production of superoxide anion radicals on the one hand and decreased antioxidant protection on the other hand (5). The resulting free radicals cause lipid peroxidation in membranes, protein oxidation, DNA mutation. Further, they induce transcription factors such as hypoxia - inducible factor alpha and nuclear factor kappa B (NF-kappaB), causing cell proliferation and hypertrophy. Increased oxidative stress in diabetic patients leads to protein oxidation and the formation of reactive carbonyl groups by direct oxidation under the influence of free radicals with the eventual formation of oxidized amino acids. Proteins and lipids can also be modified indirectly by reacting with carbonyl derivatives created by autoxidation of carbohydrates with the formation of AGE (glycation end products) and ALE (lipo-oxidation end products). Nonenzymatic reaction between glucose and the free amino groups of proteins, lipids, and nucleic acid molecules leads to the formation of molecules which no longer possess their former structural and functional properties (5).

Lipid peroxidation is the most pronounced negative phenomenon in the functioning of free radicals and is an autocatalytic, progressive and mostly irreversible process. Peroxidation of polyunsaturated fatty acids consists of three major steps: initiation, propagation and termination. Lipid peroxidation results in the formation of cytotoxic products, one of them being malondialdehyde (MDA). MDA is an accepted marker of lipid peroxidation and is used in the evaluation of oxidative stress. Highly reactive secondary products of lipid peroxidation, such as MDA, react with free SH groups and NH2 groups of amino acids, peptides, proteins, nucleotides, and phospholipids altering their functional properties. MDA reacts with proteins of blood vessels, such as collagen, changing its structure. Many studies have shown that its concentration is significantly elevated in type 1 diabetes and that optimal glycoregulation/glycemic control is necessary to reduce lipid peroxidation and MDA concentrations (6).

Diabetic nephropathy is a major chronic complication of diabetes and is one of the leading causes of end stage renal failure and mortality in diabetes. Basically, diabetic nephropathy is a glomerular disease that goes through five basic stages: glomerular hyperfiltration, normoalbuminuric phase, incipient nephropathy (microalbuminuria, urinary albumin excretion: 30 - 300 mg/24 hours), manifest proteinuria, and the final stage of renal insufficiency. The first three stages of the disease are clinically subtle, with proteinuria occurring in the fourth stage as a sign of overt nephropathy. Also, the development of diabetic nephropathy can be monitored in a clinical setting by determining urinary albumin excretion (UAE) and measuring blood pressure. The diagnosis of overt nephropathy is set based on the findings of persistent proteinuria greater than 500 mg/24 hours in the absence of urinary infection, previous intensive physical activity or other factors that may temporarily increase urinary protein excretion. However, given the importance of detecting the disease at an early stage, methods to detect the presence of incipient nephropathy and monitoring of risk factors at this stage have been developed. Having this in mind, it was established that the occurrence of microalbuminuria, i.e. UEA: 30-300 mg/24 hours, is a very reliable clinical marker of progression to proteinuria and renal insufficiency. The stage characterized by microalbuminuria was termed incipient nephropathy (7). Therefore, the detection of incipient nephropathy based on microalbuminuria is an essential part, not only in the contemporary diagnosis of nephropathy, but it also serves in the regular monitoring of patients with diabetes.

Aim

In our study group comprising 50 young people with type 1 diabetes, mean age 18.9±2.8 years, disease duration longer than five years and proven incipient diabetic nephropathy, our goal was to determine the potential impact of oxidative stress on the development of overt diabetic nephropathy.

Materials and methods

Fifty young people with type 1 diabetes, mean age 18.9±2.8 years and disease duration longer than five years, and 30 healthy children of similar age and gender, who served as controls, were included in the study. The diagnosis of diabetes mellitus type 1 was based on the American Diabetes Association (ADA) criteria. By regular screening of patients with long term diabetes, we selected 50 patients with UEA: 30-300 mg/24 hours, i.e. with incipient nephropathy. In the selected patients we investigated the level of antioxidant activity by determining the levels of SH groups, as well as the degree of oxidative stress based on MDA and AAOP levels. All patients were recruited during regular rounds during the 12 month follow-up at the Endocrinology department, Clinic of Children's Internal Diseases, Clinical Center in Niš. Patient medical data was readily available during the study period. We excluded patients whose medical records held information on relevant systemic diseases. Written informed consent was provided by all participants in the study or their parents or guardians. The patients' age ranged from 17-23.5 years, mean age 18.9±2.8 years old. The control group consisted of 30 healthy children: 13 girls and 17 boys, mean age 17.1 years, range from 14.5 years to 22.7. The blood samples were collected during the regular rounds and follow-up of patients. HbA1c was determined by standard laboratory methods.

The diagnosis of incipient nephropathy was made based on the presence of microalbuminuria, i.e. urinary albumin excretion (UAE) 30-300 mg/24 hours. Antioxidant activity was determined by measuring thiol compounds (SH groups), while the degree of lipid peroxidation by measuring malondialdehyde (MDA) and advanced oxidation protein product levels (AOPP). Oxidative injury was determined by measuring malondialdehyde (MDA) levels, being the end product of lipid peroxidation. MDA in reaction with thiobarbituric acid (TBA) at high temperatures, in an acidic medium, produces chromogen-condensation product, which was composed of 1 molecule of MDA and 2 molecules of TBA, according to the method described by Andreev (8). For the measurement of plasma MDA, a modified method using molar extinction coefficient $1.56x10^5$ M^{-1} cm^{-1} was applied, where MDA levels were expressed as μmol/l concentrations. For determining the total serum SH groups, an advanced method based on the formation of colored products was used, absorbance read at 412 nm following the addition of Ellman's reagent (5,5'-dithio-bis-(2-nitrobenzoic acid) (9). SH group levels were expressed in μmol/L.

Advanced oxidation protein products (AOPP) were determined spectrophotometrically using chloramine T solution, which in the presence of potassium iodide absorbs at 340 nm, as described by Witko-Sarsat (10).

Data are presented as means and standard deviations or medians and interquartile ranges. Testing normality of the data was performed by means of Shapiro-Wilk's test. Comparison of arithmetic means of two samples was performed by t-test or Mann-Whitney U test, depending on data distribution. Interdependence of oxidative stress biomarkers was investigated by Pearson rank correlation coefficient. Logistic regression analysis was used to test biomarkers of oxidative stress as independent risk factors for disease development. Data analysis was performed by statistical software SPSS 16.0.

Results

The study included 50 patients with diabetic nephropathy and 30 healthy subjects. The values of biomarkers of oxidative stress are shown in Table 1.

Patients had significantly higher MDA levels compared to healthy subjects ($p < 0.001$). SH concentration is significantly lower in patients than in controls ($p < 0.001$). AOPP level was significantly higher in patients than in the control group ($p < 0.001$).

Correlation analysis showed a statistically significant negative correlation between lipid peroxidation (MDA) and antioxidant defense system (SH) ($r=-0.451$, $p < 0.001$) (Figure 1), as well

Table 1. Biomarkers of oxidative stress in study groups

	Diabetic nephropathy n = 50	Control group n = 30	t/z[1]	p
MDA (μmol/L)	51.28 ± 12.76 52.10 (43.74 - 58.75)	17.54 ± 6.35[a] 17.34 (13.95 – 20.11)[b]	15.729	< 0.001
SH (μmol/L)	132.32 ± 36.60 129.40 (106.56 -154.07)	189.22 ± 42.90 183.18 (153.26-212.10)	5.402	< 0.001
AOPP (μmol/L)	48.82 ± 13.84 50.50 (37.66 - 58.47)	18.45 ± 1.73 18.86 (16.99 -19.99)	7.454	< 0.001

[a]- arithmetic mean ± standard deviation, [b] – median (interquartile range), [1] Mann-Whitney U test

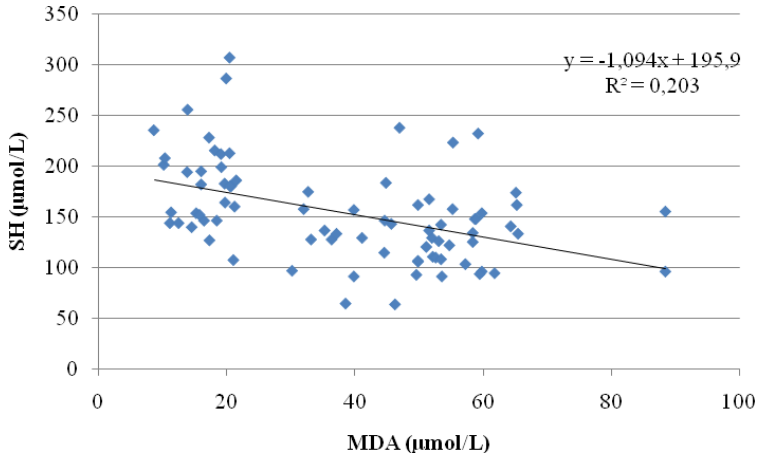

Figure 1. Correlation between MDA and SH levels

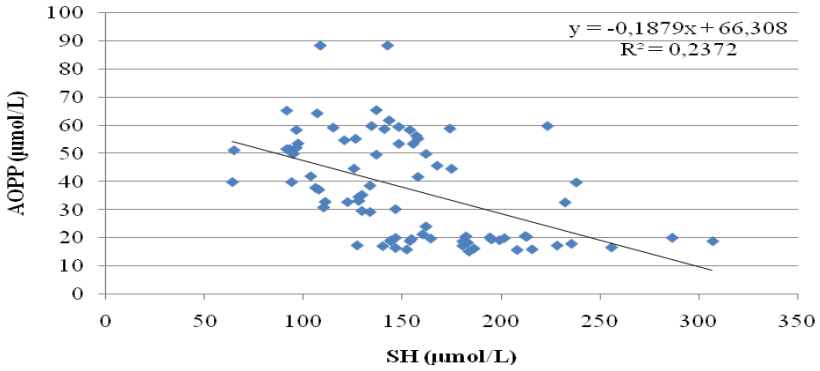

Figure 2. Correlation between AOPP and SH levels

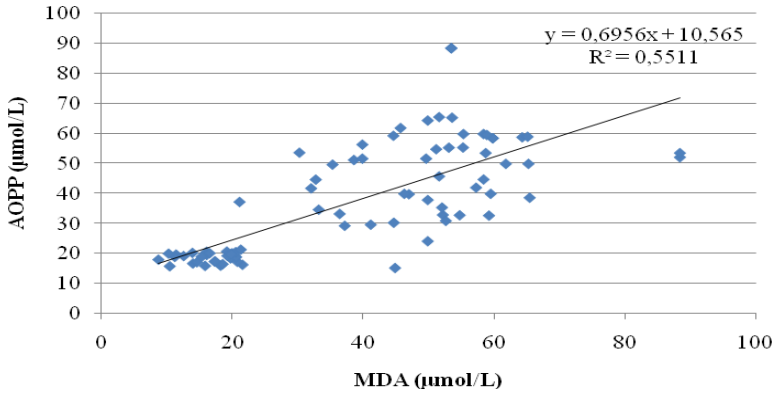

Figure 3. Correlation between MDA and AOPP concentrations

as between SH and AOPP (r=-0.487, p <0.001) (Figure 2). A statistically significant positive correlation exists between MDA and AOPP (r=0.742, p <0.001) (Figure 3) (Table 2).

Univariate logistic regression analysis showed that MDA is an independent risk factor. An increase in MDA by 1μmol increases the risk of the development of the disease by 32.4%. A reduction in SH by 1μmol/L increases the risk of the development of the disease by 4% (Table 3).

Discussion

Type 1 diabetes and its complications are typical pathological conditions in the development of which oxidative stress is involved (11).

Chronic complications of diabetes are a significant public health problem. They are a major cause of mortality in people with diabetes. The primary target for hyperglycemic damage is the endothelium of blood vessels. Four major pathways are involved in the pathogenesis of this damage. They include: increased flux through the polyol pathway, increased formation of glycation end-products, increased activation of protein kinase C and overactivity of hexosamine pathway. Although it was believed that each pathway operates separately, which is why therapeutic interventions had no effect, today it is known that the increased production of reactive oxygen species in the mitochondrial electron chain are a link between hyperglycemia and the aforementioned biochemical pathways (12). Hence, the interest in the role of oxidative stress in the development of chronic complications of diabetes, such as diabetic nephropathy, is increasing.

The main reasons for increased production of free oxidative radicals and their impact on the development of diabetes are hyperglycemia and increased production of free fatty acids. Hyperglycemia leads to non-enzymatic glycolysation of proteins causing their irreversible damage (irreversible glycosylation products), but also activates aldose reductase leading to a fall in NADPH levels, reduction in activity of glutathione reductase and antioxidant glutathione production ("glycotoxicity").Oxidation of accumulated free fatty acids produces toxic products of lipid peroxidation – malonil-dialdehyde ("lipotoxicity"). Pancreatic beta cells are usually very sensitive to the effects of reactive free radicals as the activity of antioxidant enzymes - superoxide dismutase, catalase, and es-pecially glutathione- peroxidase is primarily low in these cells (13).

Free oxidative radicals, therefore, have two major adverse effects: direct (oxidizing and damaging DNA, proteins, lipids, etc) and indirect - where radicals function as signaling molecules which activate cellular stress mechanisms via TNF-kappaB, causing direct damage to pancreatic beta cells (through TNF-α and INF-γ), and promoting development of insulin resistance (TNF-α interferes with tyrosine kinase activity and functioning of the insulin receptor). An increased production of free radicals leads to chain cleavage of DNA, interferes with mitochondrial ATP production and induces lipid peroxidation which can irreversibly damage the β-cells, leading to the development of diabetes and its complications (14).

Lipid peroxidation primarily affects cell membrane lipids (possibly pancreatic β-cell membranes) or other molecules. Intensified lipid peroxidation has been proven in type 1 diabetes and is often associated with the occurrence of early complications and atherosclerosis. Acceleration of lipid peroxidation results in the formation of toxic products, such as malondialdehyde (MDA), an accepted marker of lipid peroxidation. In our patients with long-term type 1 diabetes and diagnosed diabetic nephropathy, excessive lipid peroxidation was confirmed, associated with a statistically significant increase in the production of malondialdehyde (MDA) compared to controls ($p < 0.001$) (51.28±12.76 μmol/L vs 17.54±6.35 μmol/L). These results are consistent with data in recent reports (15). Similarly, an analysis of AOPP (advanced oxidation protein products) showed significant AOPP elevation in the diabetic nephropathy group compared to controls, and this difference attained a high level of statistical significance ($p < 0.001$) (48.82±13.84 vs 18:45± 1.73 μmol/L) which is in accordance with reports in literature where increased oxidation of plasma proteins was detected in patients with diabetic nephropathy (16). Also, a statistically significant rise in MDA serum levels accompanied by consistently low serum levels of SH group was detected in the diabetic nephropathy group ($r=-0.451$, $p < 0.001$) (Table 2). Univariate logistic regression analysis showed that unlike the relationship between MDA and SH groups, a significant relationship between SH groups and AOPP could not be determined, despite the evident rise in AOPP.

Such high levels of MDA are deemed an independent risk factor – an increase in MDA by 1μmol increases the risk of the development of the disease by 32.4%, while a reduction in SH by 1μmol/L also increases the risk of the development of the disease by 4% (Table 3). Recent data confirm the results of our study which suggest that reduced antioxidant activity and intensified lipid peroxidation are crucial for the development of complications in type 1 diabetes (17). It can be concluded that low antioxidant activity (in our study, low thiol - SH groups) and intensified lipid peroxidation (in our study a high MDA) are important in the generation of complications in diabetes type 1 such as diabetic nephropathy. An increased level of advanced oxidation protein products (AOPP) probably indicates that this mechanism is important for later occurrence of clinically manifest nephropathy with imminent renal failure but not incipient nephropathy.

Conclusion

As the primary localization of thiol compounds is in the endothelium of arteries, it can be concluded that a reduction in thiol (glutathione) levels, as a result of intensified lipid peroxidation, is a key event in the initial pathogenesis of diabetes type 1 complications, such as diabetic nephropathy.

References

1. Lenzen S. Oxidative stress: the vulnerable beta-cell. Biochem Soc Trans 2008;36(Pt 3):343-7. [CrossRef] [PubMed]
2. Negre-Salvayre A, Auge N, Ayala V, Basaga H, Boada J, Brenke R, et al. Pathological aspects of lipid peroxidation. Free Radic Res 2010;44(10): 1125-71. [CrossRef] [PubMed]
3. Valko M, Leibfritz D, Moncol J, Cronin MT, Mazur M, Telser J. Free radicals and antioxidants in normal physiological functions and human disease. Int J Biochem Cell Biol 2007;39(1):44-84. [CrossRef] [PubMed]
4. Pham-Hui LA, He H, Pham-Hui C. Free radicals, antioxidants in disease and health. Int J Biomed Sci Jun 2008;4(2):89-96. [PubMed]
5. Kulkarni R, Acharya J, Ghaskadbi S, Goel P. Thresholds of oxidative stress in newly diagnosed diabetic patients on intensive glucose-control. PLoS one 2014;9(6):e100897. [CrossRef] [PubMed]
6. Goodarzi MT, Navidi AA, Rezeai M, Babahmadi-Rezaei H. Oxidative damage to DNA and lipids: correlation with protein glycation in patients with type 1 diabetes. J Clin Lab Anal 2010;24(2):72-6. [CrossRef] [PubMed]
7. Dronavalli S, Duka I, Bakris GL. The pathogenesis of diabetic nephropathy. Nat Clin Pract Endocrinol Metab 2008;4(8):444-52 [CrossRef] [PubMed]
8. Andreeva JL, Kozemjakin AL, Kiskin A. A modification of thiobarbituturic test for measuring lipid peroxidation product. Lab Delo 1998;11:41-3.
9. Ellman LG. Tissue sulphydril groups. Arch Biochem Biophys 1959;82:70-7. [CrossRef] [PubMed]
10. Martín-Gallán P, Carrascosa A, Gussinyé M, Domínguez C. Biomarkers of diabetes-associated oxidative stress and antioxidant status in young diabetic patients with or without subclinical complications. Free Radical Biol Med 2003;34(12): 1563-74. [CrossRef] [PubMed]
11. Ceriello A. New insights on oxidative stress and diabetic complications may lead to a "causal" antioxidant therapy. Diabetes Care 2003;26(5): 1589-96. [CrossRef] [PubMed]
12. Ramakrishna V, Jailkhani R. Evaluation of oxidative stress in insulin dependent diabetes mellitus patients. Diagn Pathol 2007;2:22. [CrossRef] [PubMed]
13. Kostolanska J, Jakus V, Barak L. Glycation and lipid peroxidation in children and adolescents with type 1 diabetes mellitus with and without diabetic complications. J Pediatr Endocrinol Metab 2009;22(7):635-43. [CrossRef] [PubMed]
14. Maritim AC, Sanders RA, Watkins JB 3rd. Diabetes, oxidative stress and antioxidants: a review. Journal Biochem Toxicol 2003;17(1):24-38. [CrossRef] [PubMed]
15. Bahatia S, Shukla R, Venkata Madhu S, Kaur Gambhir J, Madhava Prabhu K. Antioxidant status, lipid peroxidation and nitric oxide end products in patients of type 2 diabetes mellitus with nephropathy. Clin Biohem 2003;36(7):557-62. [CrossRef] [PubMed]
16. Mohammedi K, Bellili Munoz N, Driss F, Roussel R, Seta N, Fumeron F, et al. Manganese superoxide dismutase (SOD2) polymorphisms, plasma advanced oxidation protein products (AOPP) concentration and risk of kidney complications in subjects with type 1 diabetes. PloS one 2014;9(5): e96916. [CrossRef] [PubMed]
17. Mohora M, Virgolici B, Paveliu F, Lixandru D, Muscurel C, Greabu M. Free radical activity in obese patients with type 2 diabetes mellitus. Rom J Inter Med 2006;44(1):69-78. [PubMed]

Originalni rad **UDK: 616.379-008.64-053.2:616.61-008.6]:616-008.9:577**
doi:10.5633/amm.2017.0315

MEHANIZMI UTICAJA OKSIDATIVNOG STRESA NA RAZVOJ DIJABETESNE NEFROPATIJE KOD TIPA 1 DIJABETESA

Jelena Vučić[1], Sandra Stanković[1], Karin Vasić[1], Tatjana Cvetković[2,3]

Klinika za dečje bolesti, Klinički centar Niš, Srbija[1]
Univerzitet u Nišu, Medicinski fakultet, Centar za biohemiju, Niš, Sbija[2]
Klinika za neurologiju, Klinički centar Niš, Srbija[3]

Kontakt: Jelena Vučić,
Ul. Studenička 34, Niš, Srbija
jvucic70@gmail.com

U seriji od 50 mladih sa dijabetesom tipa 1 srednje uzrasne dobi 18.9±2.8 godina, trajanjem bolesti dužim od 5 godina i dokazanom incipijentnom dijabetesnom nefropatijom, utvrditi potencijalni uticaj oksidativnog stresa na razvoj dijabetesne nefropatije. Dijagnoza incipijentne nefropatije postavljana je na osnovu pojave mikroalbuminurije, tj. urinarne ekskrecije albumina (UEA) 30-300 mg za 24 sata. Određivana je antioksidativna aktivnost tiolnih jedinjenja (SH grupe), intenzitet lipidne peroksidacije merenjem malondialdehida (MDA) i merenje uznapredovalih produkta oksidacije proteina (AOPP). Dokazana je insuficijentna antioksidativna zaštita sa dramatičnim padom aktivnosti tiolnih jedinjenja u grupi dijabetesne nefropatije u odnosu na kontrolu (132,32±36,60 μmol/l vs 189,22±42,90 μmol/l, p <0,001). Ovo može biti objašnjenje za intenzivirani oksidativni stres, odnosno lipidnu peroksidaciju sa povećanom produkcijom malondialdehida (MDA) u grupi dijabetesne nefropatije u odnosu na kontrolu (51,28±12,76 μmol/l vs 17,54±6,35 μmol/l, p <0,001), te povećane uznapredovale produkte oksidacije proteina (AOPP) kod obolelih u odnosu na kontrolu (48,82±13,84 vs 18,45±1,73 μmol/l, p <0,001). Korelaciona analiza je pokazala da postoji statistički značajna negativna korelacija između MDA i SH (r=-0,451, p <0,001), kao i između SH i AOPP (r=-0,487, p <0,001). Ipak, samo MDA predstavlja statistički značajan rizik za nastanak dijabetesne nefropatije. Univarijnantna logistička regresiona analiza je pokazala da je MDA nezavisni faktor rizika. Povećanje MDA za 1μmol povećava rizik za 32.4% za razvoj dijabetesne nefropatije. Smanjenje SH za 1μmol/L povećava rizik za razvoj dijabetesne nefropatije za 4%. Zaključak je da što je niža antioksidativna zaštita tiolnih jedinjenja to je intenzivnija lipidna peroksidacija (MDA), a time i veći rizik za nastanak dijabetesne nefropatije. *Acta Medica Medianae 2017;56(3):94-100.*

Ključne reči: *dijabetesna nefropatija, oksidativni stres, tiolna jedinjenja (SH), malondialdehid (MDA), uznapredovali produkti oksidacije proteina (AOPP)*