Volume 12 • Number 3 • September 2021 • em00774

JOURNAL OF CLINICAL AND EXPERIMENTAL INVESTIGATIONS

RESEARCH ARTICLE

Ischemia Modified Albumin as a Novel Biochemical Indicator in Peripheral Artery Patients

Mustafa Etli [1], *Hasan Basri Savas* [2*]

[1] Alanya Alaaddin Keykubat University, Faculty of Medicine, Department of Cardiovascular Surgery, Alanya, Antalya, Turkey

[2] Alanya Alaaddin Keykubat University, Faculty of Medicine, Department of Medical Biochemistry, Antalya, Turkey

ABSTRACT

Objective: Peripheral artery disease (PAD) is the name given to blocked blood flow in major arteries other than cerebral and coronary circulation. The diagnosis and treatment of PAD is very important because of the high frequency of PAD. In this study, it was aimed to investigate the possible change in next generation oxidative stress-antioxidant status indicators as a biochemical parameter that may be useful in the diagnosis and follow-up of PAD.

Method: Forty healthy controls and 40 male and female patients with peripheral artery disease who applied to the Alanya Alaaddin Keykubat University Research and Training Hospital were enrolled in the study on a voluntary basis. Total antioxidant status (TAS), total oxidant status (TOS), oxidative stress index (OSI), ischemia modified albumin (IMA) and Thiol balance were studied with the colorimetric method.

Results: The TOS ($p < 0.001$), OSI ($p < 0.001$), IMA ($p = 0.027$), total thiol (0.004) levels were higher in the peripheral artery patients compared to the control group, while the TAS ($p < 0.001$) and native thiol / total thiol ratio (index 3) ($p = 0.018$) values were significantly lower.

Conclusion: The increase in TOS and IMA levels may be used as an indicator of peripheral artery disease. There is an increase in inflammation in the ischemia and atherosclerosis processes. An ischemia-specific protein called ischemia-modified albumin may be used as a rapid and practical laboratory marker in common peripheral artery disease.

Keywords: peripheral arterial disease, oxidative stress, antioxidants, atherosclerosis, biomarkers

Correspondence:

Hasan Basri Savas

Address: Alanya Alaaddin Keykubat University, Faculty of Medicine, Department of Medical Biochemistry, Antalya, Turkey

Email: hasan.savas@alanya.edu.tr

Received: 20.01.2021,
Accepted: 11.03.2021
https://doi.org/10.29333/jcei/10960

INTRODUCTION

Peripheral artery disease (PAD) is the name given to the blocking of blood flow in major arteries other than cerebral and coronary circulation. PAD may occur for many reasons such as thromboembolism, vasculitis, and degeneration. However, the most common condition that causes PAD is atherosclerosis. PAD usually occurs in the lower extremities and leads to increased mortality. PAD often exhibits silent picture in its clinical course. Since PAD has silent clinical course, possible laboratory parameters that can provide early diagnosis of the disease are very valuable. PAD follows a clinical course ranging from a simple leg pain to limb loss. Patients often apply to the cardiovascular surgery clinic after repeated polyclinic admissions. Patients may apply to the clinic with severe ischemia findings from simple claudication. In order to diagnose PAD, firstly, Doppler Ultrasonography (USG), contrast-enhanced Computed Tomography (CT), Magnetic Resonance (MR) imaging, angiography, definitive diagnosis are performed by conventional angiography. The frequency of PAD in the population defined as adults reaches 32%. Over the age of 80, the frequency of PAD exceeds 40%. PAD is more common in men. The diagnosis and treatment of PAD are very important due to the high frequency of PAD, leading to increased morbidity and mortality [1-3]. Oxidative stress and antioxidant status are in equilibrium in organisms. Disruption of this balance towards oxidative stress accelerates atherosclerosis. Therefore, increased oxidative stress is likely to cause an increase in the frequency of PAD. Biochemically,

many new generation indicators of oxidative stress and antioxidant status have begun to be measured. When the increase in oxidative stress exceeds the antioxidant capacity, the amount of free oxygen radicals begins to increase. Free oxygen radicals cause various damages. They adversely affect lipid structures, membranes and genetic material. They disrupt the structure of proteins. On the other hand, there are many enzymes showing antioxidant activity. These include superoxide dismutase, glutathione peroxidase, catalase and many similar enzymes. Additionally, antioxidant components that are not in enzyme structure also affect this balance. The total antioxidant status (TAS) and total oxidant status (TOS) are general laboratory parameters that show the overall balance of oxidative stress and antioxidant status very well. Thanks to the oxidative stress index (OSI) calculated on these two parameters, it may be possible to maintain a general idea instead of measuring all the effects of oxidative damage and all components of antioxidant capacity separately [4-15]. The thiol structure contains a sulfhydryl group. Thanks to sulfhydryl, thiol is very effective in preventing oxidative stress. If there is a significant increase in oxidative stress, thiol is oxidized, and the disulfide bond is formed. If oxidative stress decreases, and antioxidant capacity increases, disulfide bonds can turn back into the thiol structure. This cycle is called the dynamic thiol balance. Dynamic thiol balance is disrupted at the end of the increase in oxidative stress. For this reason, the incidence of many diseases, including atherosclerosis and PAD, increases. Oxidation enhances ischemia. The albumin structure is modified due to oxidative stress and accompanying ischemia. The N-terminal modified albumin structure is called ischemia modified albumin (IMA) [16]. Therefore, TAS, TOS, OSI, dynamic thiol balance and IMA levels, which are next generation oxidative stress and antioxidant status indicators, are likely to be a significant laboratory marker in the diagnosis, treatment, and monitoring of PAD. The present study aimed to investigate the possible change in next generation oxidative stress-antioxidant status indicators as a biochemical parameter that could be useful in the diagnosis and follow-up of PAD.

MATERIALS AND METHODS

Design

Forty healthy controls and 40 male and female patients with peripheral artery disease who applied to the Alanya Alaaddin Keykubat University Research and Training Hospital were enrolled in the study with their voluntary consent.

Ethical Situation

The research was carried out with the approval of the Alanya Alaaddin Keykubat University Clinical Research Ethics Committee, dated 01-08-2019 and numbered 10354421-2019 / 7. During the clinical trial, the Declaration of Helsinki was followed in accordance with the ethical principles.

Measured Parameters

TAS, TOS, OSI, IMA and Thiol balance were studied by the colorimetric method after all blood was collected. The data obtained were subjected to statistical analysis, and the existence of a significant variation between the groups was checked. $P < 0.05$ was determined as the level of statistical significance.

Total Antioxidant Status, Total Oxidant Status and Oxidative Stress Index Analysis

Serum total antioxidant statuses (TAS), total oxidant statuses (TOS) measurement and Oxidative Stress Index (OSI) calculation was made according to the standard colorimetric method, which has been used in many studies before [9].

Ischemia Modified Albumin Measurement

Serum IMA values were measured with Albumin cobalt binding analysis. For determination of the amount of cobalt not bound to albumin, 25 µl of dithiothreitol (final concentration 1.67 mmol / l) was introduced to the measuring cuvette following incubation and mixed so that dithiothreitol did not bind to albumin and formed a colored complex with cobalt. The colored complex that was formed was analyzed by spectrophotometry at a wavelength of 500 nm [16].

Thiol Balance Measurement

Thiol measurement was made according to the standard colorimetric method, which has been used in many studies before [17].

RESULTS

Forty patients with peripheral artery disease and 40 healthy volunteers were enrolled in this study. Thirty patients were male, and 10 were female. The mean age of the patients was 68 ± 8 years. The characteristics of the patient and control groups are presented in **Table 1**. There was a difference between the TAS, TOS, OSI, IMA and Total thiol values of the participants in the patient group in comparison to the healthy adults. The TAS, OSI, IMA and total thiol levels of the patients were greater than those of the healthy adults, and their TAS levels were lower than those of the healthy adults. On the other hand, there was no significant difference between the groups regarding the Native thiol and Disulfide values (**Table 2**).

When the ROC analysis of the laboratory results was performed, the area under the curve was found to be the highest in OSI, TOS, total thiol and IMA, respectively. While the cut-off value for OSI was determined as 2.85, the sensitivity was 75%, specificity was 88%, PPV was 86%, and NPV was 77%. While the cut-off value for TOS was determined as 3.63, the sensitivity was 73%, specificity was 80%, PPV was 78%, and NPV was 74%. While the cut-off value for total thiol was determined as 380.92, the sensitivity was 55%, specificity was 83%, PPV was 76%, and NPV was 65%. While the cut-off value for IMA was determined as

Table 1. The characteristics of the patient and control groups

	Healthy Control (Mean±SD)	Peripheral Arterial Disease (n=40)
Age (years) (mean±SD)	66.4±7.8	68.4±8.6
Sex	Male: 28 (70%) Female: 12 (30%)	Male: 30 (75%) Female: 10 (25%)
Smoking	14 (35%)	14 (35%)
Diabetes Mellitus	16 (40%)	17 (42.5%)

Table 2. Comparison of Laboratory Results of Patients and Healthy Adults

Group	Healthy Control (Mean±SD)	Peripheral artery disease (Mean±SD)	p
TAS	1.35±0.22	1.06±0.20	0.001
TOS	3.00±0.63	4.20±1.29	0.001
OSI	2.27±0.56	4.16±1.60	0.001
IMA	0.68±0.16	0.82±0.34	0.035
Native Thiol	274.80±71.82	294.89±83.29	0.133
Total thiol	306.85±77.86	364.01±95.62	0.004
Disulfide	34.85±14.06	37.45±17.31	0.751

TAS: Total antioxidant status, TOS: Total oxidant status, OSI: Oxidative stress index, IMA: Ischemia-modified albumin

Table 3. ROC Analysis of Laboratory Results

Test Results	AUC	P	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
TAS	0.174	0.001	0.084	0.264
TOS	0.769	0.001	0.657	0.881
OSI	0.849	0.001	0.757	0.940
IMA	0.637	0.036	0.513	0.760
Native Thiol	0.598	0.133	0.471	0.724
Total Thiol	0.693	0.003	0.576	0.811
Disulfide	0.521	0.751	0.390	0.651

TAS: Total antioxidant status, TOS: Total oxidant status, OSI: Oxidative stress index, IMA: Ischemia-modified albumin, AUC: area under the curve

0.82, the sensitivity was 35%, specificity was 85%, PPV was 70%, and NPV was 57% (**Table 3**, **Figure 1**).

DISCUSSION

Peripheral artery disease is an important vascular disorder that mostly develops based on atherosclerosis and mostly involves the lower extremities. It is one of the four main topics of cardiovascular diseases. Despite the high frequency and clinical importance of the disease, angiography used in the diagnosis may only be useful in the late period [1-3]. There is a need for a practical, inexpensive and non-invasive biochemical test that can be useful in diagnosis for early PAD. It is thought that increased oxidative stress may trigger atherosclerosis and peripheral artery disease. In our study, as the mean ± standard deviation in control and peripheral artery groups, respectively, TAS was found as 1.35 ± 0.22 and 1.06 ± 0.2, TOS was found as 2.99 ± 0.63 and 4.20 ± 1.29, and OSI was found to be 2.27 ± 0.57 and 4.17 ± 1.60. There was a significant and mutually supportive change regarding all three parameters between the groups ($p<0.001$). For this reason, according to our results, the deterioration of the balance between oxidant-antioxidant status towards the increase of oxidative stress may be evaluated in relation to peripheral artery disease. The new generation oxidative stress indicator IMA levels known to increase in the case of ischemia as mean ± standard deviation in the control and peripheral artery groups were found respectively as 0.68 ± 0.17 and 0.82 ± 0.34 ($p = 0.027$). There was a significant change regarding the IMA levels between the groups. Therefore, the increase in the IMA levels may be used as an indicator of peripheral artery disease.

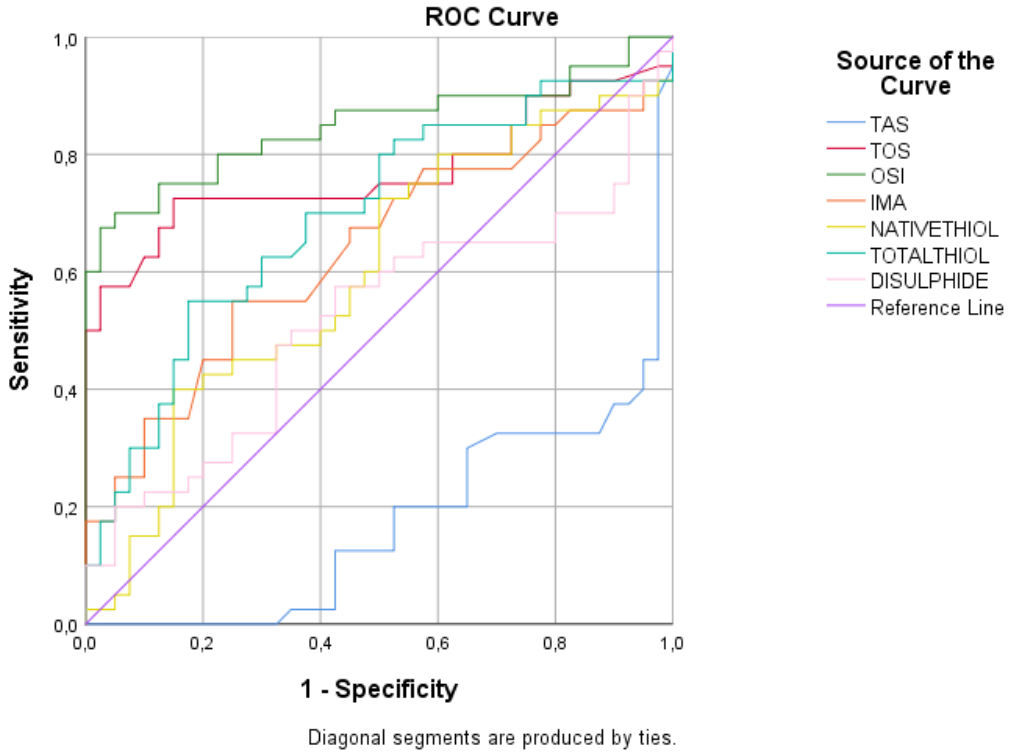

Figure 1. ROC Analysis of Laboratory Results

There is an increase in inflammation in the ischemia and atherosclerosis processes. Ischemia-specific protein called ischemia-modified albumin may be used as a rapid and practical laboratory marker in common peripheral artery disease.

IMA also occurs due to high oxidative stress in different models of ischemia that affect other organs other than the myocardium [18]. Serum IMA level may vary in end stage renal disease, cerebrovascular ischemia, non-cardiac ischemic diseases, cardiopulmonary resuscitation, acute mesenteric ischemia, pulmonary embolism, as well as peripheral vascular diseases [18-21]. In this study, the values of OSI, TOS, Total thiol and IMA increased in accordance with the literature. IMA does not rise in the case of immune system disorders, gastrointestinal diseases and non-ischemic heart diseases [18]. IMA was determined as significantly greater in patients with acute ischemic stroke, and there was a positive correlation between thiobarbituric acid-reactive substance (TBARS) and IMA levels [22]. Oxidative stress biomarkers are increased in patients with scleroderma. IMA was identified as significantly greater in patients with systemic sclerosis [23].

Plasma IMA values were greater in the acute period of rat models with mesenteric ischemia in comparison to controls [24]. Ischemia leads to proinflammatory reaction cascades causing creation of reactive oxygen species (ROS) [21-27].

In oxidative stress and tissue inflammation, the increase in blood disulfide values and disulfide / native thiol, disulfide / total thiol ratios, indicating thiol / disulfide imbalance in vitiligo, were significant [28]. As shown in this study, the rise in oxidative stress and tissue inflammation was found to be significant. In our study, OSI, TOS, total thiol, IMA supported the result because of the increase in inflammation due to oxidative stress and atherosclerosis due to ischemia in peripheral artery patients.

It was determined that the oxidant-antioxidant balance was towards oxidants in acute ischemia which was observed in patients with acute cerebral infarction (ACI) and acute intracerebral hemorrhage (AIH). The absence of a difference in albumin, IMA, TAS, TOS, IMA / albumin ratio (IMAR) and OSI values shows that comparable processes have a function in the two conditions. It was ascertained that the oxidant-antioxidant balance was already disrupted in the early hours of stroke, much before detection of ischemia on Cranial CT (CCT) images. Ischemic modified albumin / albumin ratio, TOS and OSI were found to be useful in the early diagnosis of not only ischemic but also hemorrhagic strokes. Additionally, the change between patients with acute stroke and healthy adults indicates that these biomarkers may be utilized to make a distinction between actual stroke and healthy adults mimicking stroke [29]. The authors claimed that biomarkers emerging from the deterioration of the oxidant-antioxidant balance due to ischemia could be useful in the early identification of ischemia in an acute stroke study. In our study, we think that we can detect the effects of ischemia occurring in PA patients at an early stage with oxidant-antioxidant balance parameters. Their study supported the results of our study.

IMA is useful in cardiac diseases in evaluating early ischemia before the onset of non-transient cardiac damage, preventing progression to infarction, and preventing later complications. Clinical use of IMA in cardiac ischemia is useful, sensitive, cheap, and easy in terms of early diagnosis [30]. There are similar causes such as atherosclerosis and ischemia in the pathogenesis of cardiac and peripheral events. In the study conducted in cardiac ischemia, measuring the IMA level in the early period supports our study by claiming that cardiac complications will decrease.

It is known that increased oxidative stress can lead to many serious diseases [31,32]. Our research results show that there is a possible relationship between increased oxidative stress levels and peripheral artery disease.

LIMITATIONS

This study was conducted as a clinical study on patients who were diagnosed with peripheral artery disease and had consent. It is a single center research. The number of patients included in the study was limited to 40 due to its single center. More significance levels can be reached in new studies to be conducted with larger patient groups. In addition, there was a lack of research in the literature examining serum thiol disulfide balance in peripheral artery patients. Although the results of our research fill this gap, the discussion section of our article was not as extensive as we would like due to the lack of similar research to compare.

CONCLUSION

Oxidant-antioxidant markers will contribute to prevention of a process from claudication to amputation in the extremity with early evaluation of ischemia before the onset of vascular damage in peripheral artery disease. Although oxidant-antioxidant markers were used at acute stages in most studies, and although there was slow progressing ischemia and atherosclerosis in PAD, the markers were found to be significantly higher. In patients with PAD, the use of thiol-disulfide balance ischemia-modified albumin levels, which is an indicator of oxidative stress and antioxidant status, may provide early diagnosis and treatment. In addition, after the clinical use of the parameters we investigated, the mortality, morbidity, unnecessary radiation exposure and patient costs of the patients can be significantly reduced. The clinical use of OSI, TOS, total thiol and IMA in peripheral ischemia is beneficial, sensitive, cheap, and easy for early diagnosis. There is a need for studies on oxidant-antioxidant markers in patients with peripheral artery disease and diseases related to ischemia.

HIGHLIGHTS

According to our research results:

- The increase in total oxidant status (TOS) levels may be used as an indicator of peripheral artery disease.
- The increase in ischemia modified albumin (IMA) levels may be used as an indicator of peripheral artery disease.
- There is an increase in inflammation in the ischemia and atherosclerosis processes. An ischemia-specific protein called ischemia-modified albumin may be used as a rapid and practical laboratory marker in common peripheral artery disease.

Author contributions: All authors have sufficiently contributed to the study, and agreed with the results and conclusions.

Funding: During this study, no financial or spiritual support was received neither from any pharmaceutical company that has a direct connection with the research subject, nor from a company that provides or produces medical instruments and materials which may negatively affect the evaluation process of this study.

Declaration of interest: No conflicts of interest between the authors and / or family members of the scientific and medical committee members or members of the potential conflicts of interest, counseling, expertise, working conditions, shareholding and similar situations in any firm.

Protection of humans: The authors declare that the procedures were followed according to the regulations established by the Clinical Research and Ethics Committee and to the 2013 Helsinki Declaration of the World Medical Association.

Data confidentiality: The authors declare having followed the protocols in use at their working center regarding patients' data publication.

REFERENCES

1. Velioglu Y, Yuksel A. Complete blood count parameters in peripheral arterial disease. Aging Male. 2019; 22(3): 187-91. doi: 10.1080/13685538.2019.1588873.
2. Delaney CL, Smale MK, Miller MD. Nutritional Considerations for Peripheral Arterial Disease: A Narrative Review. Nutrients. 2019; 29; 11(6): 1219. doi: 10.3390/nu11061219.
3. Berti-Hearn L, Elliott B. A closer look at lower extremity peripheral arterial disease. Nursing. 2018; 48(1): 34-41. doi: 10.1097/01.NURSE.0000527596.79097.9a.
4. Ozturk SA, Ceylan C, Serel TA, Doluoglu OG, Soyupek AS, Guzel A, et al. Protective effect of theophylline on renal functions in experimental pneumoperitoneum model. Ren Fail 2015; 37(6): 1044-9. doi: 10.3109/0886022X.2015.1040706.
5. Gumral N, Saygin M, Asci H, Uguz AC, Celik O, Doguc DK, et al. The effects of electromagnetic radiation (2450 MHz wireless devices) on the heart and blood tissue: role of melatonin. Bratisl Med J 2016; 117 (11): 665-71. doi: 10.4149/BLL_2016_128.
6. Canbolat MF, Savas HB, Gultekin F. Improved catalytic activity by catalase immobilization using c-cyclodextrin and electrospun PCL nanofibers. J. Appl. Polym. Sci. 2017; 134:4: 1-7. doi: 10.1002/app.44404.
7. Altuntaş A, Yiğit A, Uz E, İnal S, Kidir V, Aydin B, et al. The relationship between serum fetuin a levels and fetuin gene polymorphism in hemodialysis patients. Biomedical Research. 2017; 28 (2): 495-502.

8. Cankara FN, Özmen Ö, Savas HB, Şahin Y, Günaydın C. Gastroprotective effect of tarantula cubensis extract in the indomethacin-induced peptic ulcer model in rats. Acta Med. Alanya 2020; 4(3): 278-84. doi: 10.30565/medalanya.797406.
9. Savas HB, Gultekin F, Ciris İM. Positive effects of meal frequency and calorie restriction on antioxidant systems in rats. North Clin Istanbul. 2017; 4(2): 109-16. doi: 10.14744/nci.2017.21548.
10. Yiğit A, Savaş HB. Glutathione S-Transferase Enzyme Gene Polymorphisms and Cardiovascular Diseases. J Clin Anal Med. 2016; 7(5): 749-52. doi: 10.4328/JCAM.4323.
11. Türkkan A, Savas HB, Yavuz B, Yiğit A, Uz E, Bayram NA, et al. The Prophylactic Effects of Viscum Album in Streptozotocin-Induced Diabetic Rats. North Clin Ist. 2016; 3(2): 83-9. doi: 10.14744/nci.2016.22932. 2016.
12. Gultekin F, Nazıroğlu M, Savas HB, Çiğ B. Calorie restriction protects against apoptosis, mitochondrial oxidative stress and increased calcium signaling through inhibition of TRPV1 channel in the hippocampus and dorsal root ganglion of rats. Metab Brain Dis. 2018; 33(5): 1761-74. doi: 10.1007/s11011-018-0289-0.
13. Savran M, Ozmen O, Erzurumlu Y, Savas HB, Asci S, Kaynak M. The Impact of Prophylactic Lacosamide on LPS-Induced Neuroinflammation in Aged Rats. Inflammation. 2019; 42(5): 1913-24. doi: 10.1007/s10753-019-01053-7.
14. Savran M, Asci H, Ozmen O, Erzurumlu Y, Savas HB, Sonmez Y, et al. Melatonin protects the heart and endothelium against high fructose corn syrup consumption-induced cardiovascular toxicity via SIRT-1 signaling. Hum Exp Toxicol. 2019; 38(10): 1212-1223. doi: 10.1177/0960327119860188.
15. Savran M, Aslankoc R, Ozmen O, Erzurumlu Y, Savas HB, Temel EN, et al. Agomelatine could prevent brain and cerebellum injury against LPS-induced neuroinflammation in rats. Cytokine. 2019; 127: 154957. doi: 10.1016/j.cyto.2019.154957.
16. Erdem D, Savas HB, Erdem N, Kuloglu Genc S, Avcioglu G. Ischemia Modified Albumin as a New Marker for Diagnosis of Early Pregnancy Losses. Int J Acad Med Pharm 2020: 2(3): 222-7. doi: 10.29228/jamp.44816.
17. Erel O, Neselioglu S. A novel and automated assay for thiol/disulphide homeostasis. Clin Biochem. 2014; 47(18): 326-32. doi: 10.1016/j.clinbiochem.2014.09.026.
18. Sbarouni E, Georgiadou P, Voudris V. Ischemia modified albumin changes - review and clinical implications. Clin Chem Lab Med. 2011; 49(2): 177-84. doi: 10.1515/CCLM.2011.037.
19. Erdem SS, Yerlikaya FH, Çiçekler H, Gül M. Association between ischemia-modified albumin, homocysteine, vitamin B(12) and folic acid in patients with severe sepsis. Clin Chem Lab Med. 2012; 50(8): 1417-21. doi: 10.1515/cclm-2011-0794.
20. Piwowar A, Knapik-Kordecka M, Warwas M. Ischemia-modified albumin level in type 2 diabetes mellitus-Preliminary report. Dis Markers. 2008; 24(6): 311-7. doi: 10.1155/2008/784313.
21. Ellidag HY, Bulbuller N, Eren E, Abusoglu S, Akgol E, Cetiner M, et al. Ischemia-modified albumin: could it be a new oxidative stress biomarker for colorectal carcinoma? Gut Liver. 2013; 7(6): 675-80. doi: 10.5009/gnl.2013.7.6.675.
22. Seneş M, Kazan N, Coşkun O, Zengi O, Inan L, Yücel D. Oxidative and nitrosative stress in acute ischaemic stroke. Ann Clin Biochem. 2007; 44(Pt 1): 43-7. doi: 10.1258/000456307779596057.
23. Borderie D, Allanore Y, Meune C, Devaux JY, Ekindjian OG, Kahan A. High ischemia-modified albumin concentration reflects oxidative stress but not myocardial involvement in systemic sclerosis. Clin Chem. 2004; 50(11): 2190-3. doi: 10.1373/clinchem.2004.034371.
24. Gunduz A, Turkmen S, Turedi S, Mentese A, Yulug E, Ulusoy H, et al. Time-dependent variations in ischemia-modified albumin levels in mesenteric ischemia. Acad Emerg Med. 2009; 16(6): 539-43. doi: 10.1111/j.1553-2712.2009.00414.x.
25. Fidan E, Mentese A, Kavgaci H, Orem A, Fidan S, Uzun A, et al. Increased ischemia-modified albumin levels in patients with gastric cancer. Neoplasma. 2012; 59(4): 393-7. doi: 10.4149/neo_2012_051.
26. Stachowicz-Stencel T, Synakiewicz A, Owczarzak A, Sliwińska A, Aleksandrowicz-Wrona E, Lysiak-Szydowska W, et al. Ischemia-modified albumin as a biochemical marker in children with neuroblastoma and soft tissue sarcomas. J Clin Lab Anal. 2011; 25(4): 255-8. doi: 10.1002/jcla.20469.
27. Ellidag HY, Eren E, Aydin O, Akgol E, Yalcinkaya S, Sezer C, et al. Ischemia modified albumin levels and oxidative stress in patients with bladder cancer. Asian Pac J Cancer Prev. 2013; 14(5): 2759-63. doi: 10.7314/apjcp.2013.14.5.2759.
28. Üstüner P. The Role of Thiol/Disulfide Homeostasis in Vitiligo: A Novel Inflammatory Marker. İstanbul İKSST Derg. 2018; 10(1): 18-24. doi:10.5222/iksst.2018.012.
29. Atik İ, Kozacı N, Beydilli İ, Avcı M, Ellidağ H, Keşaplı M. Investigation of oxidant and antioxidant levels in patients with acute stroke in the emergency service. Am J Emerg Med. 2016; 34(12): 2379-83. doi: 10.1016/j.ajem.2016.08.062.
30. Can Ü, Yosunkaya Ş. A New Marker for Ischemia: Ischemia-modified Albumin. Koşuyolu Heart J. 2017; 20(2): 148-52. doi: 10.5578/khj.10257.
31. Seflek HN, Kalkan S, Cuce G, Kılınc I, Sozen ME. Effects of Nigella sativa oil on ovarian volume, oxidant systems, XIAP and NF-kB expression in an experimental model of diabetes. Biotech Histochem. 2019; 94(5): 325-33. doi: 10.1080/10520295.2019.1566571.

32. Cuce G, Canbaz HT, Sozen ME, Yerlikaya FH, Kalkan S. Vitamin E and selenium treatment of monocrotaline induced hepatotoxicity in rats. Biotech Histochem. 2017; 92(1): 59-67. doi: 10.1080/10520295.2016.1267798.