Scientific paper

Oxidative Brain Injury Induced by Amiodarone in Rats: Protective Effect of S-Methyl Methionine Sulfonium Chloride

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Received: 11-25-2022

Abstract

Amiodarone (AMD) is a powerful antiarrhythmic drug preferred for treatments of tachycardias. Brain can be affected negatively when some drugs are used, including antiarrhythmics. S-methyl methionine sulfonium chloride (MMSC) is a well-known sulfur containing substance and a novel powerful antioxidant. It was intended to investigate the protective effects of MMSC on amiodarone induced brain damage. Rats were divided to four groups as follows, control (given corn oil), MMSC (50 mg/kg per day), AMD (100 mg/kg per day), AMD (100 mg/kg per day) + MMSC (50 mg/kg per day). The brain glutathione and total antioxidant levels, catalase, superoxide dismutase, glutathione peroxidase, paraoxonase, and Na^+/K^+-ATPase activities were decreased, lipid peroxidation and protein carbonyl, total oxidant status, oxidative stress index and reactive oxygen species levels, myeloperoxidase, acetylcholine esterase and lactate dehydrogenase activities were increased after AMD treatment. Administration of MMSC reversed these results. We can conclude that MMSC ameliorated AMD induced brain injury probably due to its antioxidant and cell protective effect.

Keywords: amiodarone, brain, oxidative stress, S-methyl methionine sulfonium chloride, free radical

1. Introduction

Amiodarone (AMD), 2-n-Butyl-3-(3',5'-diiodo-4', N-diethylaminoethoxy-3-benzoylfuran, is an effective antiarrhythmic drug used all around the world for decades. According to the four Vaughan-Williams classification, AMD strongly belongs to class III antiarrhythmic drug (AAD),[1] although it shows all the effects of all electrophysiological characteristics of this classification. This drug inhibits myocardial potassium channels and alters the activity of fast sodium channels in heart.[2] Albeit its positive effect on arrhythmia treatment, AMD has been declared as having many toxic effects, due to its highly lipophilic nature, and in turn accumulation tendency on many organs like liver, lung, lens, skin, gingiva.[3–6] Likewise, AMD can easily cross blood brain barrier (BBB).[7] In addition to this, the cardiac and nerve systems (conductive cells and neurons) share the same histologically specialized cells, sodium-potassium channels, excitability, and conductivity.[8] This phenomenon can facilitate AADs penetrating through brain. AMD also triggers free radical formation by transforming into a radical itself via interaction with electron transport chain (ETC).[9] If the high oxygen demand of brain is considered, brain damage will be inevitable owing to its metabolic interactions.

S-methyl methionine sulfonium chloride (MMSC) is also known as Vitamin U, is a sulfur-containing derivative of the essential amino acid L-methionine. Nevertheless, it is not actually in the vitamin classification, but this substance is called as vitamin due to its vitamin-like effects.[10] It is mainly found in raw cabbage, tomatoes, spinach, and garlic.[11] Their consumption has been growing day by day following knowledge of their protective effects. MMSC has been reported to have many protective activities including antiulcer, lipid lowering, wound healing, hepatoprotective, renoprotective, and anti-thrombotic.[6,12–16] In addition, the most amazing and breath-taking attention of this sulfur containing compound is its antioxidant property, which has been proven by many researchers.[17–19]

In the current study, the protective effects of MMSC on amiodarone induced brain damage was investigated.

2. Materials and Methods

2. 1. Animals

In this study, 3.5–4.0 months old male Sprague Dawley rats were obtained from Istanbul University Experimental Medical Research and Application Institute, DETAE. The experimental procedures were approved by the local Animal Care and Use Committee of Istanbul University (with the certification number 2012/127). All the animals were fed with standard animal pellet food and tap water ad libitum.

2. 2. Experimental Design

AMD dose was chosen by considering the method of Reasor *et al.*[20] and MMSC dose was applied according to the method of Sokmen *et al.*[21]

Rats were divided randomly into 4 groups as follows: Group I; control group, which received corn oil (for 7 days and n=6). Group II; MMSC group, received MMSC at a dose of 50 mg/kg by gavage technique (for 7 days and n=7). Group III; AMD group; received AMD at a dose of 100 mg/kg by gavage technique (for 7 days and n = 8). Group IV; AMD+MMSC group; animals receiving MMSC (50 mg/kg) for 7 days 1 h prior to the administration of AMD (100 mg/kg) (n = 8). Due to reason of the increasing weight lose effect of high doses of AMD, which was indicated in the study of Reasor *et al.*[20] 100 mg/kg per day AMD dose was preferred in this study.

On the 8th day, all the animals, which were fasted overnight, then sacrificed. Brain tissues were taken from animals under anesthesia. All the tissues were homogenized with 0.9% NaCl, and all the homogenates were centrifuged. For MPO activity, the brain tissues were separately homogenized in hexadecyl trimethyl ammonium bromide (HETAB) solution (prepared in 50 mM phosphate buffer at a pH level 6.0) and then centrifuged. The supernatants were collected for the biochemical analysis and kept frozen until the experiments were done.

2. 3. Biochemical Experiments

From the supernatants, reduced glutathione (GSH) levels were determined according to the reduction reaction of Ellman's reagent via free thiol groups for producing a yellow substance with 5,5'-dithiobis (2- nitrobenzoic acid).[22] Lipid peroxidation (LPO) levels were determined with tiobarbutiric acid reaction[23]. Protein carbonyl (PC) levels were determined as measuring carbonyl levels with the reaction of 2,4-dinitrophenylhydrazine.[24]

Total antioxidant capacity (TAC) levels were determined with a reaction based on decolorization reaction of 2,2′-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid radical cation ($ABTS^{*+}$) by antioxidants[25]. The alteration of color is measured at 660 nm. Total oxidant status (TOS) levels were determined by the presence of o-dianisidine, ferric ammonium sulfate, and xylenol orange indicator for detecting the hydrogen peroxide levels at 660 nm.[26] Oxidative stress index (OSI) levels were calculated by the ration of TAC/TOS levels and the results were multiplied with 100 for expressing % ratio.[25,26] Reactive oxygen species (ROS) levels were determined with a fluorescent substance (2,7-dichloro fluorescein) and extinction/emission values were recorded.[27]

Catalase (CAT) activity was measured as considering the transformation of hydrogen peroxide to water and the alteration of absorbance was recorded at 240 nm.[28] Superoxide dismutase (SOD) activity was determined as regarding riboflavin related o-dianisidine reaction to increase the rate of photooxidation at 460 nm.[29] Glutathione peroxidase (GPx) activity was measured the transformation of GSH to GSSG by the presence of GR and NADPH at 366 nm.[30]

Paraoxonase (PON) activity was determined with the paraoxon ethyl substrate, and the absorbance alteration was recorded at 405 nm.[31] Myeloperoxidase (MPO) activity was determined by the presence of 4-aminoantipyrine, phenol and hydrogen peroxide, and the absorbance alteration was detected at 510 nm.[32] Acetylcholine esterase (AChE) reaction was determined at 405 nm using the acetylthiocholine iodide as substrate.[33] Lactate dehydrogenase (LDH) activity was measured at 340 nm using sodium pyruvate as substrate via NADH cofactor.[34] Sodium potassium ATPase (Na^+/K^+-ATPase) activity was determined according to the formation of phosphate and blue colored substance was recorded at 680 nm.[35] The protein levels were determined using the Lowry *et al.*[36] method.

2. 4. Statistical Analyses

Statistical analysis of biochemical results was performed via GraphPad Prism 6.0 (GraphPad Software, San Diego, California, USA). The values were expressed as means ± standard deviation (SD). The results were evaluated using an unpaired t-test and analysis of variance (ANOVA) followed by Tukey's multiple comparison tests. The value of $P < 0.05$ was considered statistically significant. The biochemical results were also evaluated by using Origin for performing principal component analysis (PCA).

3. Results

Brain GSH, LPO and PC levels are presented in Figure 1. AMD caused a significant decrease in GSH levels ($P < 0.05$) and significant increase in LPO and PC levels when comparison were made with control group ($P < 0.05$; $P < 0.01$, respectively). MMSC increased GSH and decreased LPO levels significantly in AMD group ($P < 0.05$; $P < 0.001$) (Figure 1).

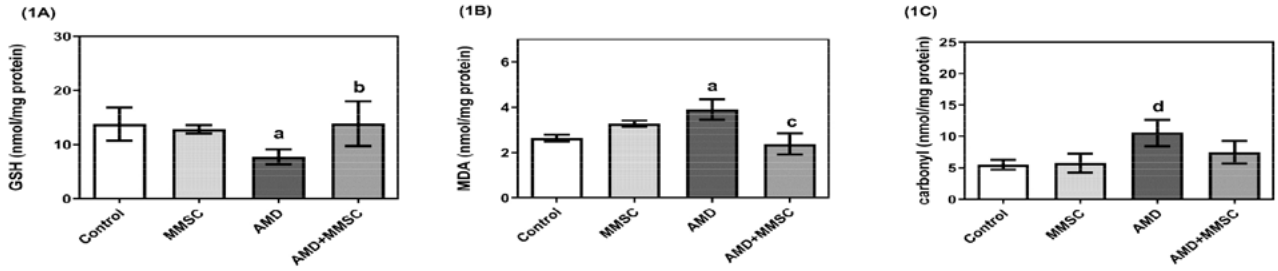

Figure 1. The brain (A) reduced glutathione (GSH), (B) lipid peroxidation (LPO) and (C) protein carbonyl (PC) levels of all groups. Each column represents mean ± SD. [a]P < 0.05 versus control group, [b]P < 0.05 versus AMD group, [c]P < 0.001 versus AMD group, [d]P < 0.01 versus control group. AMD: Amiodarone, MMSC: S-methyl methionine sulfonium chloride.

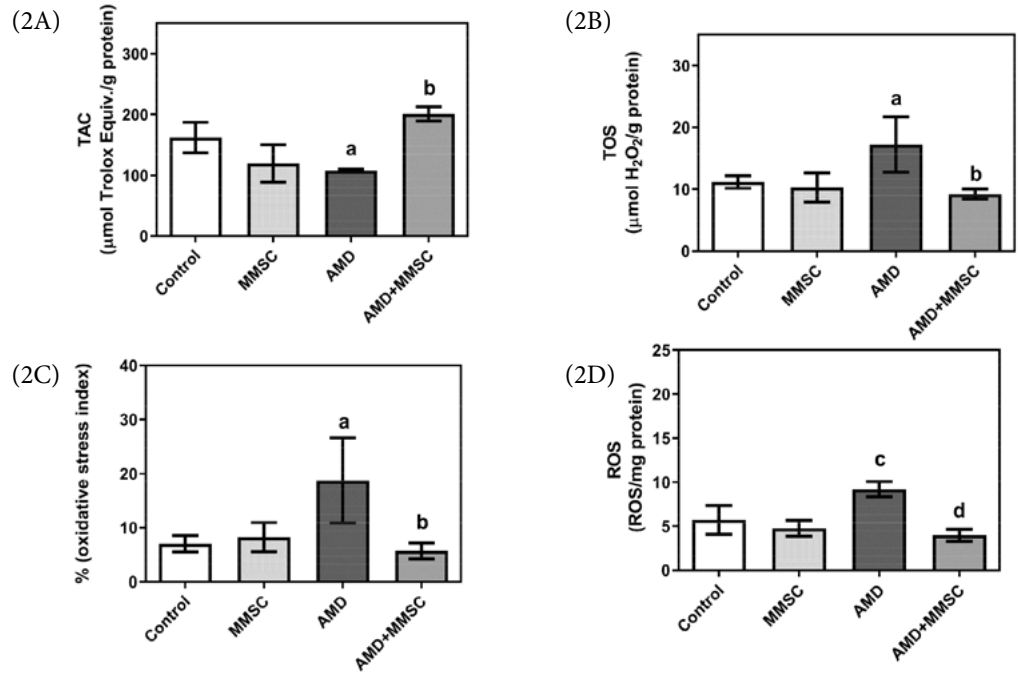

Figure 2. The brain (A) total antioxidant capacity (TAC), (B) total antioxidant status (TOS), (C) oxidative stress index (OSI) and (D) reactive oxygen species (ROS) levels of all groups. Each column represents mean ± SD. [a]P < 0.05 versus control group, [b]P < 0.01 versus AMD group, [c]P < 0.0001 versus control group, [d]P < 0.0001 versus AMD group. AMD: Amiodarone, MMSC: S-methyl methionine sulfonium chloride.

In Figure 2, brain TAC, TOS, OSI and ROS levels are shown. AMD administration decreased TAC levels significantly (P < 0.05), increased TOS and OSI (P < 0.05) significantly compared to control group. The alteration of ROS levels in AMD group was very significant as compared to control group (P < 0.0001). Administration of MMSC increased TAC

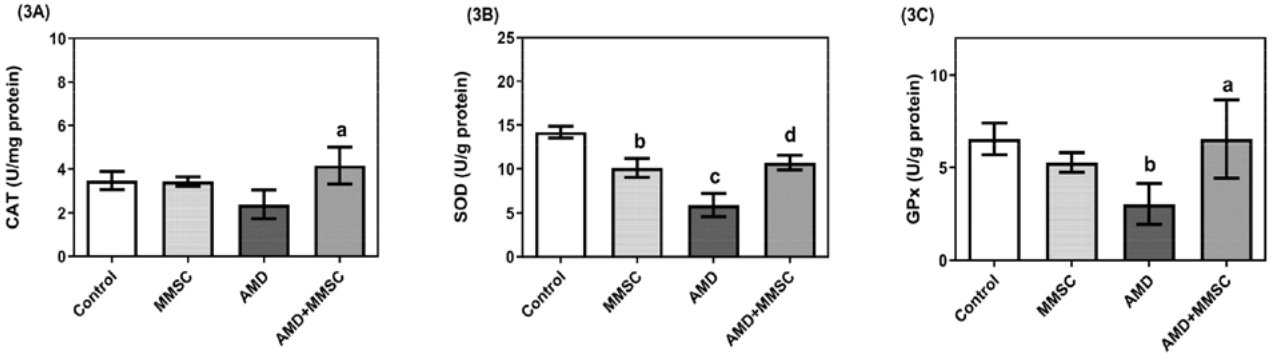

Figure 3. The brain (A) catalase (CAT), (B) superoxide dismutase (SOD) and (C) glutathione peroxidase (GPx) activities of all groups. Each column represents mean ± SD. [a]P < 0.01 versus AMD group, [b]P < 0.01 versus control group, [c]P < 0.0001 versus control group, [d]P < 0.001 versus AMD group. AMD: Amiodarone, MMSC: S-methyl methionine sulfonium chloride.

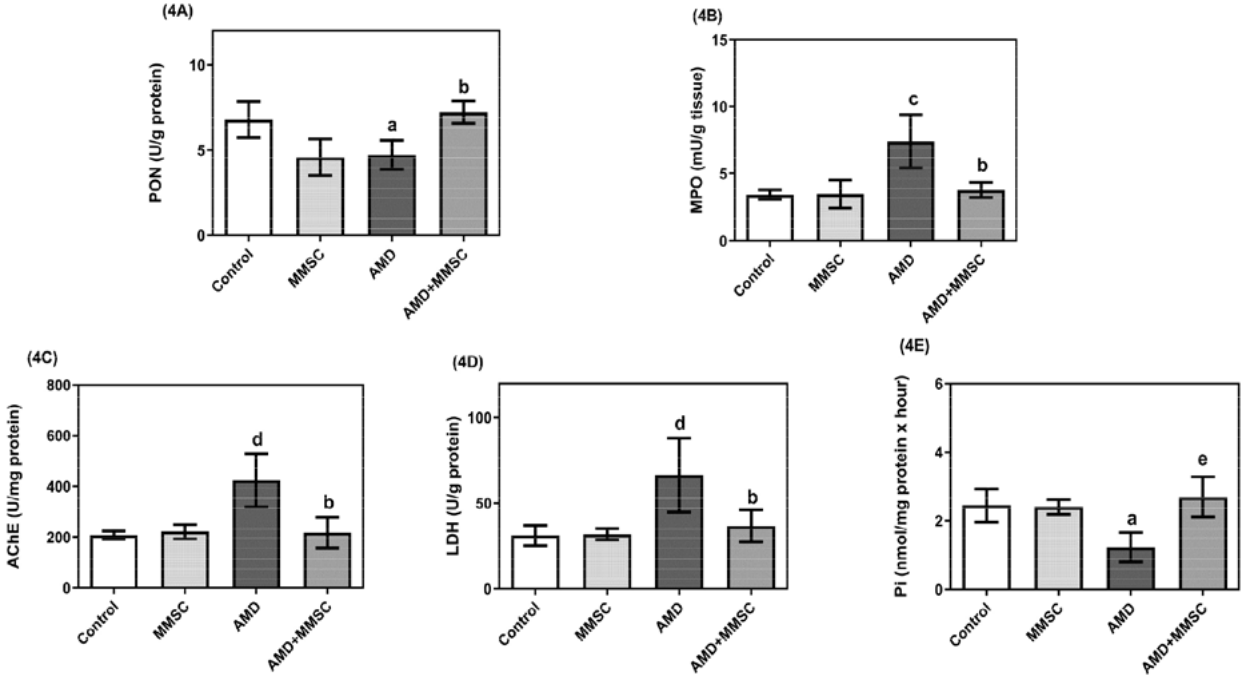

Figure 4. The brain (A) paraoxonase (PON), (B) myeloperoxidase (MPO), (C) acetylcholine esterase (AChE), (D) lactate dehydrogenase (LDH) and (E) Na^+/K^+-ATPase activities of all groups. Each column represents mean ± SD. $^aP < 0.05$ versus control group, $^bP < 0.01$ versus AMD group, $^cP < 0.01$ versus control group, $^dP < 0.001$ versus control group, $^eP < 0.001$ versus AMD group. AMD: Amiodarone, MMSC: S-methyl methionine sulfonium chloride.

levels and decreased oxidative stress parameters TOS, OSI and ROS in a significant manner ($P < 0.01$; $P < 0.0001$).

The brain CAT, SOD and GPx activities are given in Figure 3. MMSC significantly ($P < 0.01$) decreased SOD activity in comparison to control group. AMD decreased SOD and GPx activities in a significant manner when compared to control group ($P < 0.0001$; $P < 0.01$). In AMD+MMSC group, all enzyme activities showed a tendence of significant elevation as compared to AMD group ($P < 0.01$, $P < 0.001$) (Figure 3).

Brain PON, MPO, AChE, LDH and Na^+/K^+-ATPase activities of all groups are seen in Figure 4. The PON and Na^+/K^+-ATPase activities of AMD group were found to decrease ($p < 0.05$), while MPO, AChE and LDH activities were found to increase significantly as compared to control group ($p < 0.01$, $p < 0.001$, respectively). Administration of MMSC reversed these activities significantly in comparison to AMD group ($p < 0.01$, $p < 0.001$, respectively) (Figure 4).

Principal component analysis (PCA) was used to determine the correlation between all biochemical parameters (Figure 5). According to the PCA, the first two components were determined around 70.51% (as total result). PC1 and PC2 values were calculated as 61.11% and 9.40%,

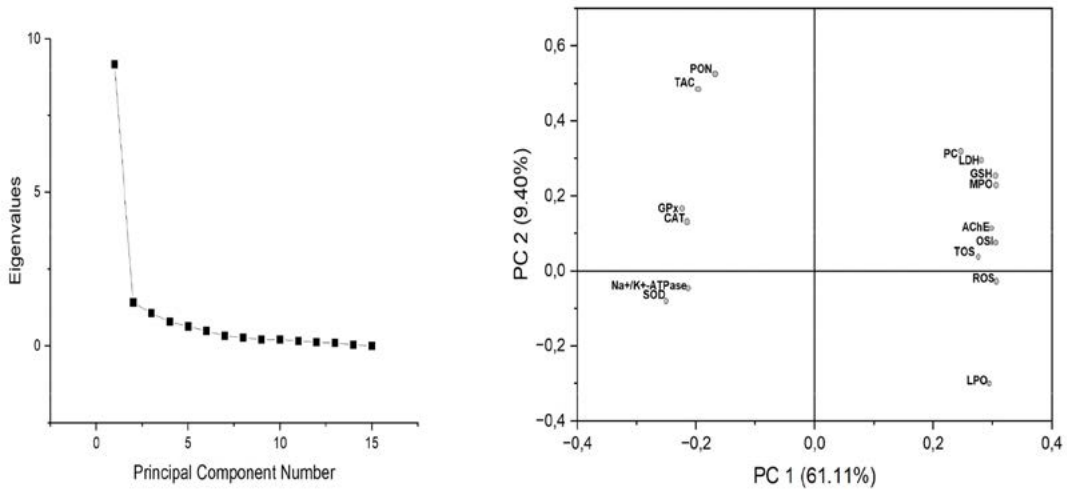

Figure 5. Principal component analysis (PCA) results for all biochemical parameters. (A) Scree plot and (B) PCA score plot of the first two PCs.

respectively. At the first component part, PC, LDH, GSH, MPO, AChE, OSI, TOS, ROS and LPO data were observed to be clustered together. These parameters were negatively correlated with PON, TAC, GPx, CAT, Na^+/K^+-ATPase and SOD (Figure 5B).

4. Discussion

The cardiac and neuronal systems share some common features due to sodium inward and potassium outward currents. Likewise, it has been reported that the cardiorespiratory system was tightly regulated with autonomic nervous system.[8] AMD is a trigger for inducing oxidative stress, and in turn ROS formation by either itself or its radical form.[9] The oxidative stress, which will occur for many reasons, may harm lipid membranes, interfere with DNA structure, and interrupt cellular respiration system.[37] By the way, oxidative stress is responsible for many brain diseases like Alzheimer's disease (AD) and Parkinson's disease, via increased protein modification and LPO levels.[38]

GSH is a vital tripeptide and shows unique antioxidant property via its sulfhydryl group which helps GSH to scavenge free radicals.[39] GSH is transformed to oxidized glutathione, GSSG. In particular, the brain ratio of these molecules must be balanced for regulating redox homeostasis and NADPH levels, as well as activities of GSH dependent enzymes like GPx.[40] When a dramatic diminishment of GSH occurs in cell media, elevated ROS is experienced. Likewise, LPO metabolisms is affected due to the existence of ROS. Elevated LPO means that there is an alteration in structure of membrane integrity and permeability.[41] In different brain disorders like Alzheimer's disease, LPO is directly associated with amyloid beta plaque formation.[42] As a free radical initiator, both AMD and other pharmacological agents like AADs directly target membrane structure by changing ion transport flux.[43] In another angle, PC products that likely occur as LPO are also unwanted threats for brain and many tissues due to oxidation of side chains of amino acids via LPO products.[44] Increased PC products change cell viability. According to the research of Zheng *et al.*,[45] most of the PC product have tendency to accumulate in mitochondria. In the present study, AMD decreased GSH levels and increased LPO and PC levels dramatically. Administration of MMSC reversed these levels in brain tissue. This ameliorating effect can be associated with membrane structure protection and antioxidant effect of MMSC, which has been published by different researchers indicating various toxicity models.[46,47]

This high oxygen demand of brain is mainly used for ATP production via oxidative phosphorylation.[48] At the end of AMD metabolism, excess iodine is released while the rest of the molecule becomes trigger for ROS formation.[49] Secondly, AMD like other cationic amphiphilic drugs, enters the cell as neutral. Thereafter, tertiary amine group is protonated because of the pH difference between inner and outer mitochondrial membrane. When protonated, AMD enters matrix, and a proton is released due to the more alkaline matrix media versus matrix intermedia. Hence, AMD accumulation begins in this way.[50] Due to these reasons, excess formation of superoxide anion will be needed to transform hydrogen peroxide (H_2O_2) by SOD, H_2O_2 will be scavenged by CAT via transforming the molecule to water and molecular oxygen.[48] Their activities were found to be decreased in AMD induced brain tissues. GPx activity was also decreased in this group, probably due to elevated levels of H_2O_2 and decreased GSH levels. An in turn, TAC levels of brain tissue dramatically decreased, and in turn TOS, OSI and ROS levels were increased. The present results are in accordance with antioxidant enzyme activities study of Hazineci *et al.*[46] on AMD induced heart damage. Sulfur containing food and other compounds like garlic, white cabbage, taurine, and N-acetyl cysteine have been reported to protect brain and other organ against damages induced by oxidative stress.[46, 51–53] MMSC reversed these activities and levels in brain tissue. Its protective effect can be explained by its unique antioxidant capacity due to its sulfur content.

PON enzymes are very sensitive during the existence of oxidative stress. In brain tissue, PON2 exist at various region, it is highly active and enhances coenzyme Q function in ETC. This helps to decrease excessive ROS production.[54] MPO catalyzes the conversion of H_2O_2 in the presence of chloride ions to hypochlorous acid (HOCl). The acid formed is capable of attacking different amino acid residues of various proteins, e.g. tyrosine residues.[55] In addition to that, HOCl may inhibit mitochondrial respiration system, thereby leading to decreased of NAD levels and distorted cellular ATP metabolism.[56] Altered PON and MPO activities have been reported in different brain disorders related to ROS elevation.[57,58] In this study, the outcomes suggest that AMD administration caused a significant diminishment of PON, and elevation of MPO activities probably due to its ability to increase ROS levels. MMSC administration reversed these activities compared to AMD group probably by successfully decreasing ROS levels.

Acetyl choline (ACh), an important neurotransmitter, is degraded by AChE. The activity of this enzyme is related to various neurologic problems like Alzheimer's disease[59], as well as age related oxidative stress and in turn, memory and learning problems. These problems may also occur by increasing AChE activity, and the excess degradation of ACh.[60] In the present study, AMD increased AChE activity as compared to control group. MMSC decreased AChE activity of AMD group as compared to AMD group. This effect of MMSC on this enzyme can be related to its sulfur group. The anti-AChE activity of sulfur groups has been published by Osmaniye *et al.*[61]

LDH is a key enzyme for glycolysis. Its activity is important for brain NADH/NAD^+ transformation, a vital marker for brain redox balance.[62] AMD has been reported

to increase LDH activity in various pulmonary and hepatic toxicity models, by promoting cell death.[63,64] This elevation has been associated with altered mitochondrial capacity and NADH/NAD^+ ratio. In this study, increased activity of LDH in brain tissues of AMD group was observed. The situation caused by AMD was reversed by MMSC probably due to its antioxidant activity.

Na^+/K^+-ATPase is an essential membrane bound enzyme. It stimulates Na^+/K^+ inward/outward movements through ATP hydrolysis. Harris *et al.*[65] indicates that this enzyme accounts for half of the total consumption of ATP in healthy brains. Its activity is also vital for protecting the membrane potential.[66] In many neurological diseases, there is a well-known connection between ROS and oxidized products, LDH activity, affected antioxidant enzyme metabolism and diminished Na^+/K^+-ATPase activity.[67] In the present study, diminished activities of this enzyme in AMD treated group was observed. This diminishment may be either due to ROS-triggering effect and metabolic alterations caused by AMD. Gray *et al.*[68] and Pitt *et al.*[69] revealed that AMD inhibited this enzyme in cardiac tissue. The present findings are coherent with the functional relationship between cardiovascular system and nervous system as Borowicz and Banach[8] mentioned. MMSC administration increased this activity in AMD+MMSC group as compared to AMD group. This effect may be due to membrane repairing and antioxidant effects of MMSC, as earlier indicated by Rácz *et al.*,[47] Turkyilmaz and Yanardag,[70] and Topaloglu et al.,[18] respectively.

Our biochemical results for antioxidant and other toxicity parameters were proven to be in accordance with performing correlation analysis with PCA. These results showed that the elevations of toxicity parameters and, diminishments of antioxidant levels and enzyme activities were evidence for existence of conditions formed by AMD.

5. Conclusion

To summarize, the present finding proves that the antioxidant property of MMSC (an important sulfur containing substance) had excellent scavenging effect on ROS and protected redox balance by reversing the deleterious effects of AMD in brain tissue.

Data Availability Statement

The author declares that [the/all other] data supporting the findings of this study are available within the article.

6. References

1. S. A. Papiris, C. Triantafillidou, L. Kolilekas, D. Markoulaki, E. D. Manali, *Drug Saf.* **2010**, *33*, 539–558. **DOI:**10.2165/11532320-000000000-00000
2. R. M. Colunga Biancatelli, V. Congedo, L. Calvosa, M. Ciacciarelli, A. Polidoro, L. Iuliano, *J. Geriatr. Cardiol.* **2019**, *16*, 552–566. **DOI:**10.1155/2019/3418950
3. K. Jaworski, I. Walecka, L. Rudnicka, M. Gnatowski, D. A. Kosior, *Med. Sci. Monit.* **2014**, *20*, 2369–2372. **DOI:**10.12659/MSM.890881
4. L. T. Keng, M. T. Liao, *Postgrad. Med. J.* **2018**, *94*, 603. **DOI:**10.1136/postgradmedj-2018-135779
5. U. Turk, B. G. Turk, S. G. Yılmaz, E. Tuncer, E. Alioğlu, T. Dereli, *Middle East Afr. J. Ophthalmol.* **2015**, *22*, 258–260. **DOI:**10.4103/0974-9233.154411
6. I. B. Turkyilmaz, R. Yanardag, *Bulg. Chem. Commun.* **2019**, *51*, 20–24.
7. M. Banach, M. Popławska, K. K. Borowicz-Reutt, *Epilepsy Res.* **2018**, *140*, 105–110. **DOI:**10.1016/j.eplepsyres.2018.01.003
8. K. K. Borowicz, M. Banach, *Pharmacol. Rep.* **2014**, *66*, 545–551. **DOI:**10.1016/j.pharep.2014.03.009
9. A. C. Nicolescu, J. L. Comeau, B. C. Hill, L. L. Bedard, T. Takahashi, J. F. Brien, W. J. Racz, T. E. Massey, *Toxicol. Appl. Pharmacol.* **2007**, *220*, 60–71. **DOI:**10.1016/j.taap.2006.12.031
10. E. H. Choi, S. B. Lee, D. Y. Lee, G. T. Kim, S. M. Shim, T. S. Park, *Nutrients*. **2019**, *11*, 1334. **DOI:**10.3390/nu11061334
11. N. R. Augspurger, C. S. Scherer, T. A. Garrow, D. H. Baker, *J. Nutr.* **2005**, *135*, 1712–1717. **DOI:**10.1093/jn/135.7.1712
12. S. Gezginci-Oktayoglu, I. B. Turkyilmaz, M. Ercin, R. Yanardag, S. Bolkent, *Protoplasma*. **2016**, *253*, 127–135. **DOI:**10.1007/s00709-015-0796-3
13. W. S. Kim, Y. J. Yang, H. G. Min, M. G. Song, J. S. Lee, K. Y. Park, J. J. Kim, J. H. Sung, J. S. Choi, H. J. Cha, *Pharmacology*. **2010**, *85*, 68–76. **DOI:**10.1159/000276495
14. A. S. Salim, *J. Pharm. Sci.* **1992**, *81*, 70–73. **DOI:**10.1002/jps.2600810114
15. S. Sancar Bas, I. B. Turkyilmaz, S. Bolkent, R. Yanardag, *Eur. J. Biol.* **2016**, *75*, 1–10.
16. K. Seri, T. Matsuo, T, Taniguchi, K. Amemiya, M. Kudo, G. Saito, T. Kato, *Arzneimittelforschung*. **1980**, *30*, 1694–1703.
17. S. Tunali, S. Kahraman, R. Yanardag, *Hum. Exp. Tox.* **2015**, *34*, 904–910. **DOI:**10.1177/0960327114561665
18. D. Topaloglu, I. B. Turkyilmaz, R. Yanardag, *J. Biochem. Mol. Toxicol.* **2022**, *36*, e23124. **DOI:**10.1002/jbt.23124
19. D. Mahmarzayeva, B. B. Bayrak, I. B. Turkyilmaz, O. Sacan, R. Yanardag, *J. Biochem. Mol. Toxicol.* **2022**, *36*, e23126. **DOI:**10.1002/jbt.23126
20. M. J. Reasor, C. M. McCloud, T. L. Beard, D. C. Ebert, S. Kacew, M. F. Gardner, K. A. Aldern, K. Y. Hostetler, *Toxicology.* **1996**, *106*, 139–147. **DOI:**10.1016/0300-483X(95)03175-F
21. B. B. Sokmen, S. Tunali, R. Yanardag, *Food Chem. Toxicol.* **2012**, *50*, 3562–3566. **DOI:**10.1016/j.fct.2012.07.056
22. E. Beutler, Glutathione in red cell metabolism, A Manual of Biochemical Methods, Grune and Stratton, Eds. New York, **1975**, 112–114.
23. A. Ledwozyw, J. Michalak, A. Stepień, A. Kadziołka, *Clin. Chim. Acta.* **1986**, *155*, 275–283. **DOI:**10.1016/0009-8981(86)90247-0
24. R. L. Levine, D. Garland, C. N. Oliver, A. Amici, I. Climent, A. G. Lenz, B. W. Ahn, S. Shaltiel, E. R. Stadtman, *Methods*

Enzymol. **1990**, *186*, 464–478. **DOI:**10.1016/0076-6879(90)86141-H
25. O. Erel, *Clin. Biochem.* **2004**, *37*, 277–285. **DOI:**10.1016/j.clinbiochem.2003.11.015
26. O. Erel, *Clin. Biochem.* **2005**, *38*, 1103–1111. **DOI:**10.1016/j.clinbiochem.2005.08.008
27. Y. Zhang, J. Chen, H. Ji, Z. G. Xiao, P. Shen, L. H. Xu, *BMC Complement. Altern. Med.* **2018**, *18*, 343. **DOI:**10.1186/s12906-018-2414-3
28. H. Aebi, *Methods Enzymol.* **1984**, *105*, 121–126. **DOI:**10.1016/S0076-6879(84)05016-3
29. A. A. Mylroie, H. Collins, C. Umbles, J. Kyle, *Toxicol. Appl. Pharmacol.* **1986**, *82*, 512–520. **DOI:**10.1016/0041-008X(86)90286-3
30. A. Wendel, *Methods Enzymol.* **1981**, *77*, 325–333. **DOI:**10.1016/S0076-6879(81)77046-0
31. C. E. Furlong, R. J. Richter, S. L. Seidel, A. G. Motulsky, *Am. J. Hum. Genet.* **1988**, *43*, 230–238.
32. H. Wei, K. Frenkel, *Cancer Res.* **1991**, *51*, 4443–4449.
33. G. L. Ellman, K. D. Courtney, V. Andres, R. M. Feather-Stone, *Biochem. Pharmacol.* **1961**, *7*, 88–95. **DOI:**10.1016/0006-2952(61)90145-9
34. R. Bais, M. Philcox, *Eur. J. Clin. Chem. Clin. Biochem.* **1994**, *32*, 639–655.
35. A. Ridderstap, S. Bonting, *Am. J. Physiol.* **1969**, *217*, 1721–1727. **DOI:**10.1152/ajplegacy.1969.217.6.1721
36. O. H. Lowry, N. J. Rosebrough, A. L. Farr, R. J. Randall, *J. Biol. Chem.* **1951**, *193*, 265–275. **DOI:**10.1016/S0021-9258(19)52451-6
37. M. Smeyne, R. J. Smeyne, *Free Radic. Biol. Med.* **2013**, *62*, 13–25. **DOI:**10.1016/j.freeradbiomed.2013.05.001
38. P. Poprac, K. Jomova, M. Simunkova, V. Kollar, C. J. Rhodes, M. Valko, *Trends Pharmacol. Sci.* **2017**, *38*, 592–607. **DOI:**10.1016/j.tips.2017.04.005
39. D. Shen, T. P. Dalton, D. W. Nebert, H. G. Shertzer, *J. Biol. Chem.* **2005**, *280*, 25305–25312. **DOI:**10.1074/jbc.M500095200
40. D. Dwivedi, K. Megha, R. Mishra, P. K. Mandal, *Neurochem. Res.* **2020**, *45*, 1461–1480. **DOI:**10.1007/s11064-020-03030-1
41. R. M. Lasagni Vitar, C. G. Reides, S. M. Ferreira, S. F. Llesuy, *Food Funct.* **2014**, *5*, 557–563. **DOI:**10.1039/c3fo60392j
42. D. A. Butterfield, *Ageing Res. Rev.* **2020**, *64*, 101049. **DOI:**10.1016/j.arr.2020.101049
43. C. L. H. Huang, L. Wu, K. Jeevaratnam, M. Lei, *J. Cardiovasc. Electrophysiol.* **2020**, *31*, 579–592. **DOI:**10.1111/jce.14347
44. R. Fan, L. M. Schrott, S. Snelling, J. Felty, D. Graham, P. L. McGauly, T. Arnold, N. L. Korneeva, *BMC Neurosci.* **2020**, *21*, 4. **DOI:**10.1186/s12868-020-0552-2
45. J. Zheng, C. L. Hu, K. L. Shanley, O. A. Bizzozero, *Neurochem. Res.* **2018**, *43*, 609–618. **DOI:**10.1007/s11064-017-2456-9
46. H. Hazineci, İ. B. Türkyılmaz, Ü. V. Üstündağ, E. Emekli-Alturfan, B. Alev Tüzüner, H. İpekçi, T. Tunalı-Akbay, R. Yanardağ, A. Yarat, *Experimed.* **2020**, *10*, 1–6. **DOI:**10.26650/experimed.2020.0004
47. I. Rácz, E. Páldi, G. Szalai, T. Janda, M. Pál, D. Lásztity, *J. Plant Physiol.* **2008**, *165*, 1483–1490. **DOI:**10.1016/j.jplph.2006.03.020
48. L. E. Salminen, R. H. Paul, *Rev. Neurosci.* **2014**, *25*, 805–819.
49. R. A. Eid, M. S. A. Zaki, M. Al-Shraim, M. A. Eldeen, M. A. Haidara, *Ultrastruct. Pathol.* **2021**, *45*, 49–58. **DOI:**10.1080/01913123.2020.1864076
50. A. C. Nicolescu, Y. Ji, J. L. Comeau, B. C. Hill, T. Takahashi, J. F. Brien, W. J. Racz, T. E. Massey, *Toxicol. Appl. Pharmacol.* **2008**, *227*, 370–379. **DOI:**10.1016/j.taap.2007.12.009
51. E. Akbay, B. Erdem, A. Ünlü, A. B. Durukan, M. A. Onur, *Kardiochir. Torakochirurgia Pol.* **2019**, *16*, 88–92. **DOI:**10.5114/kitp.2019.86361
52. X. Niu, S. Zheng, H. Liu, S. Li, *Mol. Med. Rep.* **2018**, *18*, 4516–4522.
53. H. Song, J. Cui, V. V. Mossine, C. M. Greenlief, K. Fritsche, G. Y. Sun, Z. Gu, *Exp. Ther. Med.* **2020**, *19*, 1554–1559.
54. J. K. Blackburn, D. W. Curry, A. N. Thomsen, R. H. Roth, J. D. Elsworth, *Exp. Neurol.* **2020**, *327*, 113234. **DOI:**10.1016/j.expneurol.2020.113234
55. S. Gellhaar, D. Sunnemark, H. Eriksson, L. Olson, D. Galter, *Cell Tissue Res.* **2017**, *369*, 445–454. **DOI:**10.1007/s00441-017-2626-8
56. R. S. Ray, A. Katyal, *Neurosci. Biobehav. Rev.* **2016**, *68*, 611–620. **DOI:**10.1016/j.neubiorev.2016.06.031
57. I. Bednarz-Misa, I. Berdowska, M. Zboch, B. Misiak, B. Zieliński, S. Płaczkowska, M. Fleszar, J. Wiśniewski, A. Gamian, M. Krzystek-Korpacka, *Adv. Clin. Exp. Med.* **2020**, *29,* 71–78. **DOI:**10.17219/acem/111377
58. C. Shao, J. Yuan, Y. Liu, Y. Qin, X. Wang, J. Gu, G. Chen, B. Zhang, H. K. Liu, J. Zhao, H. L. Zhu, Y. Qian, *Proc. Natl. Acad. Sci. USA.* **2020**, *117*, 10155–10164. **DOI:**10.1073/pnas.1917946117
59. M. Simunkova, S. H. Alwasel, I. M. Alhazza, K. Jomova, V. Kollar, M. Rusko, M. Valko, *Arch. Toxicol.* **2019**, *93*, 2491–2513. **DOI:**10.1007/s00204-019-02538-y
60. S. Asha Devi, *Sci. World J.* **2009**, *9*, 366–372. **DOI:**10.1100/tsw.2009.46
61. D. Osmaniye, B. N. Sağlık, U. Acar Çevik, S. Levent, B. Kaya Çavuşoğlu, Y. Özkay, Z. A. Kaplancıklı, G. Turan, *Molecules.* **2019**, *24*, 2392. **DOI:**10.3390/molecules24132392
62. S. Cerdán, T. B. Rodrigues, A. Sierra, M. Benito, L. L. Fonseca, C. P. Fonseca, M. L. García-Martín, *Neurochem. Int.* **2006**, *48*, 523–530. **DOI:**10.1016/j.neuint.2005.12.036
63. J. W. Card, W. J. Racz, J. F. Brien, S. B. Margolin, T. E. Massey, *Toxicol. Sci.* **2003**, *75*, 169–180.
64. K. M. Waldhauser, M. Török, H. R. Ha, U. Thomet, D. Konrad, K. Brecht, F. Follath, S. Krähenbühl, *J. Pharmacol. Exp. Ther.* **2006**, *319*, 1413–1423. **DOI:**10.1124/jpet.106.108993
65. J. J. Harris, R. Jolivet, D. Attwell, *Neuron.* **2012**, *75*, 762–777. **DOI:**10.1016/j.neuron.2012.08.019
66. A. N. Shrivastava, A. Triller, R. Melki, *Neuropharmacology.* **2020**, *169*, 107461. **DOI:**10.1016/j.neuropharm.2018.12.008
67. L. N. Zhang, Y. J. Sun, S. Pan, J. X. Li, Y. E. Qu, Y. Li, Y. L. Wang, Z. B. Gao, *Fundam. Clin. Pharmacol.* **2013**, *27*, 96–103. **DOI:**10.1111/fcp.12000
68. D. F. Gray, A. S. Mihailidou, P. S. Hansen, K. A. Buhagiar, N. L. Bewick, H. H. Rasmussen, D. W. Whalley, *J. Pharmacol. Exp. Ther.* **1998**, *284*, 75–82.

69. A. D. Pitt, C. Fernandes, N. L. Bewick, P. D. Hemsworth, K. A. Buhagiar, P. S. Hansen, H. H. Rasmussen, L. Delbridge, D. W. Whalley, *Cardiovasc. Res.* **2003**, *57*, 101–108.

70. I. B. Turkyilmaz, R. Yanardag, *Marmara Pharm. J.* **2016**, *20*, 131–136. **DOI:**10.12991/mpj.20162062716

Povzetek

Amiodaron (AMD) je močna antiaritmična učinkovina, ki je primerna za zdravljenje tahikardij. Pri uporabi nekaterih učinkovin, vključno z antiartimiki, lahko pride do negativnega vpliva na možgane. S-metil metionin sulfonijev klorid (MMSC) je znana spojina, ki vsebuje žveplo, in predstavlja nov močan antioksidant. Namen raziskave je bil raziskati zaščitne učinke MMSC na poškodbe možganov, ki jih povzroča amiodaron. Podgane so bile razdeljene v naslednje štiri skupine: kontrolna skupina (s koruznim oljem), MMSC (50 mg/kg na dan), AMD (100 mg/kg na dan), AMD (100 mg/kg na dan) + MMSC (50 mg/kg na dan). Po zdravljenju z AMD so se v možganih zmanjšale ravni glutationa, in celokupnih antioksidantov, katalaze, superoksidne dismutaze, glutation peroksidaze, paraoksonaze in Na+/K+-ATPaze, povečale pa so se lipidna peroksidacija in proteinski karbonil, skupni oksidativni status, indeks oksidativnega stresa in reaktivne kisikove zvrsti, ter aktivnosti mieloperoksidaze, acetilholin esteraze in laktat dehidrogenaze. Aplikacija MMSC je te rezultate spremenila. Zaključimo lahko, da je MMSC ublažila možganske poškodbe, ki jih je povzročil AMD, verjetno zaradi antioksidativnega in zaščitnega učinka na celice.

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