Neurological Sciences (2019) 40:385–391
https://doi.org/10.1007/s10072-018-3660-3

ORIGINAL ARTICLE

Relationship between thiol-disulphide homeostasis and visual evoked potentials in patients with multiple sclerosis

Gönül Vural[1] · Şadiye Gümüşyayla[1] · Orhan Deniz[1] · Salim Neşelioğlu[2] · Özcan Erel[2]

Received: 3 June 2018 / Accepted: 23 November 2018 / Published online: 1 December 2018

Abstract

Purpose To examine the thiol-disulphide homeostasis during an optic neuritis episode in patients with multiple sclerosis and the relationship between this homeostasis and P100 wave latency.

Materials and method Visual evoked potential reviews of multiple sclerosis patients who presented with an optic neuritis episode were conducted and P100 latencies were measured. Peripheral blood samples were collected from all patients. Native thiol and total thiol concentrations were measured with the automated method that was recently developed. Their amount of disulphide bonds, disulphide/native thiol, disulphide/total thiol and native thiol/total thiol ratios were calculated. The relationship between P100 latency and thiol-disulphide homeostasis was investigated.

Results A significant positive correlation was determined between the disulphide/native thiol ratio and both mean P100 latency and maximum P100 latency ($p = 0.021$, $r = 0.136$; $p = 0.030$, $r = 0.177$, respectively).

Discussion As the balance of the plasma dominated by antioxidants moves towards the oxidant side, in other words as a higher rate of thiol is oxidised from the thiol pool, P100 latency is extended. *N*-acetylcysteine and alpha lipoic acid as well as thiol supplements can improve the thiol-disulphide balance, reinforce antioxidant defence and it can help in slowing down the demyelinating damage.

Keywords Multiple sclerosis · Optic neuritis · Thiol-disulphide homeostasis · Oxidative stress · Visual evoked potential

Introduction

Multiple sclerosis (MS) is a demyelinating disease of the central nervous system (CNS) with a chronic inflammatory immune mediation. Various pathophysiological mechanisms such as inflammation, oxidative damage, excitotoxicity, demyelination, remyelination and axonal damage are responsible for the pathogenesis of MS [1]. Oxidative stress contributes to the degeneration of oligodendroglia and destruction of the myelin; therefore, it plays a vital role in the development of the disease. Inflammation leads to oxidative stress; oxidative stress leads to inflammation and causes demyelination [2]. Therefore, clinical signs and symptoms of MS are reflections of this pathological process where inflammation and demyelination are observed [3]. Optic neuritis is one of the most prevalent clinical presentations of MS at the onset, and it is commonly involved during its course [4]. Myelin of the optic nerve, which is considered an extension of the brain, is produced by oligodendrocytes, and these cells are vulnerable to oxidative damage [5].

Oxidative stress is described as the impairment of the balance between reactive oxygen species (ROS) radicals and antioxidant molecules in favour of ROS. This imbalance leads to the impairment of the integrity of the blood-brain barrier, myelin destruction and neurodegeneration [6]. Thiol-disulphide homeostasis is very important because it allows evaluation of the redox imbalance. Thiols are antioxidant buffers. Thiols contain the –SH group. These –SH groups react with reactive oxidant molecules and reduce them, and as a result of this, they protect the organism from oxidative damage. Disulphide bonds that are produced during this reaction can be reduced to thiol groups again, and in this way, thiol-disulphide homeostasis, which has an important role in antioxidant defence, intracellular signal transmission, regulation of enzymatic

✉ Gönül Vural
gonulvrl@gmail.com

[1] Department of Neurology, Faculty of Medicine, Yıldırım Beyazıt University, 06800 Ankara, Turkey

[2] Department of Clinical Biochemistry, Faculty of Medicine, Yıldırım Beyazıt University, Ankara, Turkey

activities, detoxification and apoptosis, can be maintained [7–9]. The evaluation of this mechanism gives us more insight into the oxidative status in the plasma in contrast to the separate measurement of oxidant and antioxidant molecules.

Oxidative stress plays a vital role in relapsing-remitting multiple sclerosis (RRMS), and it occurs before the inflammatory response during relapse. MS patients develop antioxidant insufficiency and global oxidative status [10].

In this study, we examined the thiol-disulphide homeostasis during optic neuritis episodes in patients with RRMS and the relationship between this homeostasis and P100 wave latency.

Material and method

Design

RRMS patients who are already being followed and who presented with an episode of optic neuritis during the course of the disease and patients who have been diagnosed with RRMS after experiencing an episode of optic neuritis have been included in the study. Patients whose initial findings pointed to optic neuritis episodes, but did not fulfil the criteria for an MS diagnosis yet were evaluated as clinically isolated syndromes, were excluded from the study. Visual evoked potential (VEP) reviews of all patients were conducted at the electrophysiology laboratory through monocular recording with pattern reversal evoking. Investigation was made in a dimly lit and quiet room. Patients were seated 1 m away from the monitor. Stimuli were presented as a checkerboard pattern of black and white squares at a reversal frequency of 1.5 Hz. Needle electrodes were inserted into the scalp in the midline over the occipital region 2.5 cm above the inion (Oz: active electrode) and over the frontal region (Fz: reference). The ground electrode was placed on the forearm. During uninterrupted stimulation 200 responses were averaged using a portable evoked potential machine (Keypoint; Alpine Biomed). P100 wave latencies were recorded for both eyes separately. The more longer latency of P100 waves obtained for the right and left eye was recorded as the P100 maximum, provided that it is correlated with the symptom, and their average was recorded as the P100 average. For patients who had previously had an episode of optic neuritis, P100 wave latency on the side of the new symptomatic side was recorded as the P100 maximum. In light of the anamnesis and examination findings of the patients, the durations of the disease, total number of attacks that they developed and their expanded disability status scale (EDSS) scores were recorded. Venous blood samples were collected after 12 h of fasting prior to the start of the pulse steroid treatment, and these were centrifuged at 1500*g* for 10 min immediately after. Serum thiol-disulphide homeostasis was identified with the new automated measurement method developed by Erel and Neşelioğlu [7]. In this method, firstly dynamic and degradable disulphide bonds (–S–S) were reduced to free functional thiol groups (–SH) by sodium borohydride (NaBH4). Reductant NaBH4 was not utilised, and the remaining amount was consumed with formaldehyde and removed from the environment. Following this, all thiol groups containing both reduced and native thiols entered into a reaction with DTNB [5,5′-dithiobis-(2-nitrobenzoic) acid]. Native thiol (–SH) and total thiol (–SH+–S–S) levels were measured. Half of the difference between total and native thiol contents that were measured provided us with the amount of dynamic disulphide band. Following this, disulphide/native thiol (–S–S/–SH), disulphide/total thiol (–S–S/–SH+–S–S) and native thiol/total thiol (–SH/–SH+–S–S) ratios were calculated. The study was approved by the local ethics committee and informed consent was obtained from all individual participants included in the study.

Statistical analysis

The SPPS 20 (IBM Corp. Released 2011. IBM SPSS Statistics for Windows, Version 20.0. Armonk, NY: IBM Corp.) statistical package program was used to analyse the data. Mean ± standard deviation, median, minimum-maximum and percentage values were used for the variables. The homogeneity of the variances, which is one of the pre-requisites of parametric tests, was checked with the "Levene" test. The "Shapiro-Wilk" test was used to check the assumption of normality. The relationship between two continuous variables was evaluated with Pearson's correlation coefficient test, and when they did not fulfil the pre-requisites of parametric tests, the Spearman's rank correlation coefficient was used to evaluate them. The statistical significance level was accepted as $p < 0.05$ and $p < 0.01$.

Results

Eighty-six patients aged between 18 and 57 (mean age 33.5 ± 9.2) where 30 patients were male (35%) and 56 patients were female (65%) were included in the study. Clinical properties of the patients are shown in Table 1.

Native thiol, disulphide, total thiol levels and disulphide/native thiol, disulphide/total thiol, native thiol/total thiol ratios of the patients and both mean P100 latency and maximum P100 latency were correlated for analysis (Table 2). A significant positive correlation was determined between the disulphide/native thiol ratio and both mean P100 latency and maximum P100 latency ($p = 0.021$, $r = 0.136$; $p = 0.030$, $r = 0.177$, respectively). None of the parameters of thiol-disulphide homeostasis were found to be associated with age, sex, duration of disease, total number of attacks and the number of optic neuritis episodes. Just as with the P100 latency, the disulphide/native thiol ratio was positively correlated with the EDSS score ($p = 0.026$, $r = 0.240$).

Table 1 Clinical characteristics of patients

Variables	Data
Age	33 (18–57) median (min-max) 33.5 ± 9.2 mean ± SD
Gender	*n* (%)
Male	30 (35%)
Female	56 (65%)
First clinical presentation	*n* (%)
Monosymptomatic onset	33 (38%)
Polysymptomatic onset	53 (62%)
Initial symptoms	*n* (%)
Optic	45 (52%)
Sensorial	48 (56%)
Motor	34 (39%)
Cerebellar	16 (18%)
Brainstem	11 (12%)
Disease duration (years)	2 (2–21) median (min-max) 3.6 ± 3.8 mean ± SD
Total number of attack	3 (1–10) median (min-max) 3.4 ± 2.5 mean ± SD
Number of optic neuritis episodes	1 (1–3) median (min-max) 1.2 ± 0.5 mean ± SD
Expanded disability status scale-EDSS score	2(0–6.5) median (min-max) 1.9 ± 1.4 mean ± SD
Therapy	*n* (%)
Interferon	36 (42%)
Teriflunomide	7 (8.1%)
Dimethyl fumarate	2 (2.3%)
Fingolimod	14 (16.2%)
Natalizumab	3 (3.4%)
No treatment	24 (28%)
Received pulse steroid therapy	2(0–10) median (min-max) 2.8 ± 2.2 mean ± SD
Thiol-disulphide homeostasis	Mean ± SD
Native thiol (–SH), μmol/L	454.8 ± 60.7
Total thiol (–SH+–S–S), μmol/L	493.8 ± 63.7
Dynamic disulphide (–S–S), μmol/L	19.4 ± 7.9
–S–S–/–SH	0.042 ± 0.018
–S–S–/(–SH+–S–S–)	0.040 ± 0.016
–SH/(–SH+–S–S–)	0.921 ± 0.030
Visual evoked potential –P100 latency	Mean ± SD
Maximum P100 latency msn	124.3 ± 17.6
Mean P100 latency msn	119.6 ± 16.8

The results are given as number (percentage), median (min-max), mean ± standard deviation

Discussion

MS is an inflammatory, autoimmune disease that progresses with the death of oligodendrocytes, demyelination followed by axonal damage and finally loss of neurons [11]. During the course of the disease, immune cells that are active produce ROS, which causes oxidative stress and therefore contributes to demyelination, axonal damage and inflammation processes [12, 13]. The evidence obtained demonstrates that oxidative stress plays a role in demyelination [14–16].

ROS plays a key role in myelin phagocytosis. The imbalance between ROS and antioxidant capacity in favour of ROS leads to the occurrence of oxidative stress. This imbalance that shifts in favour of ROS stimulates the adhesion of monocytes onto vascular endothelium, which changes the permeability of the blood-brain barrier, and white blood cells are extravasated into the central nervous system. This inflammatory response leads to an increase in ROS production. As a result, oxidative stress causes inflammation and inflammation causes oxidative stress. Oxidative damage occurs before inflammation. Therefore, oxidative damage is the preliminary preparer of demyelination and neurodegeneration [6, 10, 12, 13, 17].

High oxygen consumption, low antioxidant levels and a high concentration of polyunsaturated fatty acids that are considerably sensitive to pre-oxidation make the CNS especially vulnerable to oxidative damage. Oxidised phospholipids produced as a result of the peroxidation of membrane lipids induce the adhesion of monocytes onto vascular endothelium [16, 18]. As another function of ageing, iron accumulation occurs in the brain, and this iron catalyses oxidative damage through Fenton's reaction and the cellular damage that is associated with this [16, 19]. On the other hand, oxidative stress impairs the function of mitochondria and their transportation to the synaptic region, causing reduced synaptic function that results in neurodegeneration [20]. Mitochondrial dysfunction plays a special role in inflammatory processes. In case of mitochondrial dysfunction, excessive production of toxic ROS is observed [21]. It plays an important role in the loss of neurons and myelin and oligodendrocytes that are harmful to the glia [22, 23].

A series of studies investigating the role played by oxidative stress in MS has been conducted, and it has been demonstrated that an increase in oxidative stress results in reduced antioxidants [6, 24–28]. In patients with MS, serum lipid peroxidation [26], plasma fluorescent lipid peroxidation products [24], cholesterol ester hydroperoxides, which are an indicator of lipid peroxidation [29], thiobarbituric acid products [25], malondialdehyde and 4-hydroxyalkenes [6] and conjugated diene [27] levels increase, which point to the presence of oxidative status. Syburra et al. [28] have shown that lipid oxidation in patients with MS resulted in a decrease in the levels of peripheral antioxidants including vitamin E, glutathione peroxidase and ubiquinone. In the same manner, Besler [25, 26] and Miller [30] have found that the total antioxidant capacity has decreased in patients with MS. In their study where they evaluated peripheral oxidative stress in patients with RRMS, Tasset et al. [10] discovered that carbonyl protein, 8-hydroxy-2′-deoxyguanocine (8OHdG), total glutathione, reduced

Table 2 The relationship between native thiol and total thiol levels, amount of dynamic disulphide bond, (–S–S–/–SH), –S–S–/(–SH+–S–S–) and –SH/(–SH+–S–S–) ratios of patients with MS in the phase of relapse and their P100 latency (mean and maximum) and other clinical features

$n = 86$		Age	Gender	Disease duration	Total number of attacks	Number of optic neuritis episodes	EDSS score	P100 latency (mean)	P100 latency (max)
Native thiol (–SH)	r	−0.110	−0.080	−0.063	0.034	−0.039	−0.052	−0.077	−0.092
	p	0.314	0.463	0.563	0.756	0.724	0.635	0.479	0.402
Total thiol (–SH+–S–S)	r	−0.144	−0.119	−0.050	0.060	−0.007	−0.005	−0.034	−0.040
	p	0.186	0.274	0.651	0.581	0.950	0.967	0.755	0.715
Dynamic disulphide (–S–S)	r	−0.162	−0.169	0.043	0.113	0.132	0.178	0.075	0.116
	p	0.136	0.119	0.693	0.299	0.227	0.100	0.491	0.286
–S–S–/–SH	r	−0.093	−0.130	0.072	0.135	0.169	0.240*	0.136*	0.177*
	p	0.397	0.231	0.512	0.215	0.120	0.026*	0.021*	0.030*
–S–S–/(–SH+–S–S–)	r	−0.098	−0.108	0.072	0.120	0.108	0.182	0.112	0.151
	p	0.370	0.323	0.510	0.271	0.324	0.094	0.303	0.165
–SH/(–SH+–S–S–)	r	0.111	0.108	−0.070	−0.120	−0.140	−0.202	−0.115	−0.154
	p	0.309	0.323	0.523	0.272	0.199	0.062	0.290	0.156

The r value is correlation coefficient. *$p < 0.05$

glutathione (GSH), GSH/oxidised glutathione (GSSG) ratio, superoxide dismutase (SOD), glutathione reductase (GRd) and global oxidative stress were significantly higher than the healthy group, whereas total antioxidant capacity, GSSG, glutathione peroxidase (GPx) and glutathione-S-transferase (GST) were lower. In other words, they revealed total antioxidant capacity that was reduced with high oxidative stress markers, and these findings show that oxidative status is dominant in MS patients. Fiorini et al. [14] discovered that the levels of oxidative modified proteins increased during relapse in RRMS patients. Oliveira et al. [31] found that the thiol levels decreased in MS patients in their study. Ljubisavljevic [32] found that advanced oxidation protein products (AOPP) increased in patients having an acute attack compared to the controls, and thiols decreased. Pasquali et al. [33] showed that AOPP levels were significantly higher than the control group, whereas thiol levels were lower in their study, conducted with 60 patients with MS in total where 30 were RRMS and 30 of the participants were SPMS patients. Karlik et al. [17] discovered that AOPP and "thiobarbituric acid reacting substances (TBARS)", which are the lipid peroxidation markers, increased both in the saliva and in the plasma compared to the controls, and they found that the total antioxidant capacity decreased.

Reactive oxygen radicals impair the permeability of the blood-brain barrier, and they expose the protected CNS tissue to the pathological effect of immune cells, leading to demyelination and axonal loss in the end [34]. The clinical manifestation of this is increased functional losses and disability. However, conducted studies could not reveal a consistent relationship between oxidative status and clinical manifestations of the disease and disability. Pasquali et al. [33] did not determine a correlation between increased AOPP levels and reduced thiol levels and duration of the disease, sleepiness, fatigue and disability in MS patients. Tasset [10] did not find a relationship between oxidative stress biomarkers and EDSS score, whereas Ljubisavljevic [32] showed that increased AOPP levels and decreased thiol levels occur concomitantly with higher disability levels. In the study conducted by Olivera et al. [31], the authors did not discover a relationship between thiol levels and disability. Karlik et al. [17] did not find a correlation between the oxidative stress markers that had increased and EDSS in MS patients.

There are two important issues here. Firstly, all of these studies conducted until now measured individual or total pro-oxidant and antioxidant parameters separately. However, this does not provide us with accurate information about the oxidative status of the plasma. This is because oxidative status is related to the imbalance between the oxidants and antioxidants rather than the decrease or increase in their quantities. Therefore, evaluating oxidative status as thiol-disulphide homeostasis can provide us with more useful information. Thiols are major antioxidants and they are major targets for ROS. –SH is a biochemical marker for oxidation reduction reactions [35]. As evaluated in the series of studies mentioned above, the assessment of this balance by evaluating both native thiol and disulphide bonds, or in other words, the entire thiol pool with the new measurement method developed by Erel et al. [7] rather than the measurement of the thiol pool only, can provide us with a better idea about oxidative status. The thiol-disulphide balance allows an objective evaluation of the dynamic redox system of the organism. The second issue is that

evoked potential reviews demonstrate the real-time status of the physiological system [36], and they can provide more clear information than total disability, which is calculated based on the damage caused by oxidative stress and demyelination.

Development of an acute inflammatory lesion in the optic nerve leads to an optic neuritis episode characterised by visual impairment. The proinflammatory cytokines and nitric oxide in the optic nerve lesion are thought to be the major determinants of full or partial signal blocks responsible for typical visual loss of optic neuritis along with demyelination. Longitudinal studies conducted with patients who develop an optic neuritis episode have shown that the acute or persistent optic nerve demyelination is associated with the increased vulnerability of the axons, and it predicts the development of axonal loss. Both the absence of myelin support and mitochondrial dysfunction, which are a result of oxidative stress, contribute to the degeneration of the demyelinated axon [37].

Lipoic acid is a natural antioxidant, and Michael Dietrich et al. [38] have shown that the mice undergoing prophylactic lipoic acid treatment in experimental autoimmune encephalomyelitis-optic neuritis models experienced increased GSH levels in their brains, and their retinal ganglion cells were preserved while degeneration decreased.

The results of our study point out that there is a positive correlation between the disulphide/native thiol ratio and P100 latency during an optic neuritis episode. Normally, as the balance of the plasma that is dominated by antioxidants shifts towards the oxidant side or more thiols are oxidised from the thiol pool, P100 latency increases. In another study that we conducted with MS patients in relapse and remission, we showed that the patients in relapse had increased disulphide levels and disulphide/native thiol ratios, or in other words, balance had shifted to the oxidative side [39]. These results show that the redox balance system shifts to the oxidative side during relapse and supports the information that increased oxidative status causes inflammation and demyelination.

On the other hand, the disulphide/native thiol ratio showed a positive correlation with EDSS score. This, independently of the relapses, may be a reflection of an ongoing inflammation and oxidative status.

As it has an important role in MS throughout the course of the disease, oxidative stress is extremely vital. It initiates inflammatory processes during the acute phase and maintains neurodegeneration during the chronic phase. MS patients develop a global oxidative status. Therefore, the use of antioxidants is promising for a better prognosis. MS patients can benefit from antioxidant supplements [40].

Need to say, the impairment of this balance is of course not specific to MS. As a matter of fact, many recent studies on this homeostasis have shown that in neurodegenerative diseases, this balance deteriorates [41–45]. The impairment of this balance might be a non-specific common pathway in various pathologies.

Our study has some limitations. Firstly, a relatively small group of patients who admitted to a single centre were included in the study. Secondly, there was no longitudinal follow-up for the patients to determine whether alteration of this homeostasis by recurrent optic neuritis episodes and axonal loss.

Conclusion

According to our knowledge, this is the first study to examine the relationship between VEP and thiol-disulphide homeostasis. Oxidative stress plays a role in MS. Under oxidative stress, the thiol-disulphide balance is impaired in favour of disulphides. The evaluation of this balance provides more useful information in comparison to the evaluation of oxidant and antioxidant molecules separately. In this study, we discovered a correlation between the shift in thiol-disulphide balance in favour of the disulphide during an optic neuritis episode with increased P100 latency.

Therefore, *N*-acetylcysteine and alpha lipoic acid and thiol supplements can correct the thiol-disulphide imbalance, reinforce antioxidant defence and helps in slowing down demyelinising damage.

Acknowledgements The biochemical analysis of the study was performed at the biochemistry lab of Ataturk Education and Research Hospital. The authors would like to express their thanks to the staff of the biochemistry lab and Özcan Erel's research assistants for the invaluable assistance, as well as MAT Statistical Analysis Service that conducted the statistical analysis of the study.

Compliance with ethical standards

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Conflict of interest The authors declare that they have no conflicts of interest.

References

1. Tully M, Shi R (2013) New insights in the pathogenesis of multiple sclerosis—role of acrolein in neuronal and myelin damage. Int J Mol Sci 14:20037–20047
2. Ortiz GG, Pacheco-Moises FP, Bitzer-Quintero OK, Ramírez-Anguiano AC, Flores-Alvarado LJ, Ramírez-Ramírez V, Macias-Islas MA, Torres-Sánchez ED (2013) Immunology and oxidative stress in multiple sclerosis: clinical and basic approach. Clin Dev Immunol 2013:1–14

3. Francis GS, Duquette P, Antel JP (1996) Inflammatory demyelinating diseases of the central nervous system. In: Bradley WG, Daroff RB, Fenickel GM, Marsden CD (eds) Neurology in clinical practice, 2nd edn. Butterworth-Heinemann, Boston, pp 1307–1343
4. Kale N (2016) Optic neuritis as an early sign of multiple sclerosis. Eye Brain 26(8):195–202
5. Apple DJ, Rabb MF (1991) Ocular pathology, 4th edn. Mosby Year Book, St. Louis
6. Ortiz GG, Macias-Islas MA, Pacheco-Moises FP, Cruz-Ramos JA, Sustersik S, Barba EA, Aguayo A (2009) Oxidative stress is increased in serum from Mexican patients with relapsing-remitting multiple sclerosis. Dis Markers 26:35–39
7. Erel O, Neselioglu S (2014) A novel and automated assay for thiol/disulphide homeostasis. Clin Biochem 47:326–332
8. Circu ML, Aw TY (2010) Reactive oxygen species, cellular redox systems, and apoptosis. Free Radic Biol Med 48:749–762
9. Biswas S, Chida AS, Rahman I (2006) Redox modifications of protein-thiols: emerging roles in cell signaling. Biochem Pharmacol 71:551–564
10. Tassed I, Agüera E, Sanchez-Lopez F, Feijóo M, Giraldo AI, Cruz AH, Gascón F, Túnez I (2012) Peripheral oxidative stress in relapsing-remitting multiple sclerosis. Clin Biochem 45:440–444
11. Compston A, Coles A (2008) Multiple sclerosis. Lancet 372:1502–1517
12. Witherick J, Wilkins A, Scolding N, Kemp K (2011) Mechanisms of oxidative damage in multiple sclerosis and a cell therapy approach to treatment. Autoimmune Dis 2011:164608
13. vanHorssen J, Witte ME, Schreibelt G, de Vries HE (2011) Radical changes in multiple sclerosis pathogenesis. Biochim Biophys Acta 1812:141–150
14. Fiorini A, Koudriavtseva T, Bucaj E, Coccia R, Foppoli C, Giorgi A, Schininà ME, DiDomenico F, De Marco F, Perluigi M (2013) Involvement of oxidative stress in occurrence of relapses in multiple sclerosis: the spectrum of oxidatively modified serum proteins detected by proteomics and redox proteomics analysis. PLoS One 8(6):e65184
15. Gonsette RE (2008) Neurodegeneration in multiplesclerosis: the role of oxidative stress and excitotoxicity. J Neurol Sci 274(1):48–53
16. Miller E, Walczak A, Saluk J, Ponczek MB, Majsterek I (2012) Oxidative modification of patient's plasma proteins and its role in pathogenesis of multiple sclerosis. Clin Biochem 45(1–2):26–30
17. Karlik M, Valokovic P, Hancinova V, Krizova L, Tothova L, Celec P (2015) Markers of oxidative stress in plasma and saliva in patients with multiple sclerosis. Clin Biochem 48:24–28
18. Usatyuk PV, Natarajan V (2012) Hydroxy alkenals and oxidized phospholipids modulation of endothelial cytoskeleton, focal adhesion and adherens junction proteins in regulating endothelial barrier function. Microvasc Res 83(1):45–55
19. Lassman H, van Horssen J (2011) The molecular basis of neurodegeneration in multiple sclerosis. FEBS Lett 585:3715–3723
20. Ansari MA, Roberts KN, Scheff SW (2008) Oxidative stress and modification of synaptic proteins in hippocampus after traumatic brain injury. Free Radic Biol Med 45:443–452
21. Miller E, Mrowicka M, Saluk-Juszczak J, Ireneusz M (2011) The level of isoprostanes as a non-invasive marker for in vivo lipid peroxidation in secondary progressive multiple sclerosis. Neurochem Res 36(6):1012–1016
22. Beckman JS, Koppenol WH (1996) Nitricoxide, superoxide, and peroxynitrite: the good, the bad, and ugly. Am J Phys 271:1424–1437
23. Friese MA, Schattling B, Fugger L (2014) Mechanisms of neurodegeneration and axonal dysfunction in multiple sclerosis. Nat Rev Neurol 10(4):225–238
24. Koch M, Mostert J, Arutjunyan AV, Stepanov M, Teelken A, Heersema D, De Keyser J (2007) Plasma lipid peroxidation and progression of disability in multiple sclerosis. Eur J Neurol 14:529–533
25. Besler HT, Comoğlu S, Okçu Z (2002) Serum levels of antioxidant vitamins and lipid peroxidation in multiple sclerosis. Nutr Neurosci 5:215–220
26. Besler HT, Comoğlu S (2003) Lipoprotein oxidation, plasma total antioxidant capacity and homocysteine level in patients with multiple sclerosis. Nutr Neurosci 6:189–196
27. Koch M, Ramsaransing GS, Arutjunyan AV, Stepanov M, Teelken A, Heersema DJ, De Keyser J (2006) Oxidative stress in serum and peripheral blood leukocytes in patients with different disease courses of multiple sclerosis. J Neurol 253:483–487
28. Syburra C, Passi S (1999) Oxidative stress in patients with multiple sclerosis. Ukr Biokhim Zh 71:112–115
29. Ferretti G, Bacchetti T, Principi F, DiLudovico F, Viti B, Angeleri VA, Danni M, Provinciali L (2005) Increased levels of lipid hydroperoxides in plasma of patients with multiple sclerosis: a relationship with paraoxonase activity. MultScler 11:677–682
30. Miller E, Mrowicka M, Malinowska K, Zolynski K, Kedziora J (2010) Effects of the whole-body cryotherapy on a total antioxidative status and activities of some antioxidative enzymes in blood of patients with multiple sclerosis-preliminary study. J Med Investig 57:168–173
31. Oliveira SR, Kallaur AP, Simão AN, Morimoto HK, Lopes J, Panis C, Petenucci DL, da Silva E, Cecchini R, Kaimen-Maciel DR, Reiche EM (2012) Oxidative stress in multiple sclerosis patients in clinical remission: association with the expanded disability status scale. J Neurol Sci 321(1–2):49–53
32. Ljubisavljevic S, Stojanovic I, Vojinovic S, Stojanov D, Stojanovic S, Cvetkovic T, Savic D, Pavlovic D (2013) The patients with clinically isolated syndrome and relapsing remitting multiple sclerosis show different levels of advanced protein oxidation products and total thiol content in plasma and CSF. Neurochem Int 62(7):988–997
33. Pasquali L, Pecori C, Lucchesi C, LoGerfo A, Iudice A, Siciliano G, Bonuccelli U (2015) Plasmatic oxidative stress biomarkers in multiple sclerosis: relation with clinical and demographic characteristics. Clin Biochem 48:19–23
34. Mirshafiey A (2007) Venom therapy in multiple sclerosis. Neuropharmacology 53:353–361
35. Calabrese V, Bella R, Testa D, Spadaro F, Scrofani A, Rizza V, Pennisi G (1998) Increased cerebrospinal fluid and plasma levels of ultra weak chemiluminescence are associated with changes in the thiol pool and lipid soluble fluorescence in MS: the pathogenic role of oxidative stress. Drugs Exp Clin Res 24:125–131
36. Fuhr P, Kappos L (2001) Evoked potentials for evaluation of multiple sclerosis. Clin Neurophysiol 112:2185–2189
37. Thompson AJ, Baranzini SE, Geurts J, Hemmer B, Ciccarelli O (2018) Multiple sclerosis. Lancet 391:1622–1636
38. Dietrich M, Helling N, Hilla A, Heskamp A, Issberner A, Hildebrandt T, Kohne Z, Küry P, Berndt C, Aktas O, Fischer D, Hartung HP, Albrecht P (2018) Early alpha-lipoic acid therapy protects from degeneration of the inner retinal layers and vision loss in an experimental auto immune encephalomyelitis-optic neuritis model. J Neuroinflammation 15(71):1–13
39. Vural G, Gumusyayla S, Bektas H, Deniz O, Ergin M, Erel O (2016) Dynamic thiol-disulphide homeostasis in patients with multiple sclerosis. WJNS 6:214–219
40. Adamczyk B, Niedziela N, Adamczyk-Sowa M (2017) Novel approaches of oxidative stress mechanisms in the multiple sclerosis pathophysiology and therapy. In: Zagon IS, McLaughlin PJ (eds) Multiple sclerosis: perspectives in treatment and pathogenesis. Codon Publications, Brisbane, pp 155–172
41. Elmas B, Erel Ö, Ersavaş D, Yürümez Y (2017) Thiol/disulfide homeostasis as a novel indicator of oxidative stress in children with simple febrile seizures. Neurol Sci 38(11):1969–1975

42. Kurt ANC, Aydın A, Demir H, Erel Ö (2017) Headache in children and dynamic thiol/disulfide balance evaluation with a new method. Neurol Sci 38(8):1495–1499
43. Vural G, Bektas H, Gumusyayla S, Deniz O, Alışık M, Erel O (2018) Impaired thiol-disulphide homeostasis in patients with axonal polyneuropathy. Neurol Res 40(3):166–172
44. Gumusyayla S, Vural G, Bektas H, Neselioglu S, Deniz O, Erel O (2016) A novel oxidative stress marker in migraine patients: dynamic thiol-disulphide homeostasis. Neurol Sci 37(8):1311–1317
45. Vural G, Gumusyayla S, Bektas H, Deniz O, Alışık M, Erel O (2017) Impairment of dynamic thiol-disulphide homeostasis in patients with idiopathic Parkinson's disease and its relationship with clinical stage of disease. Clin Neurol Neurosurg 153:50–55