Oxidative and Antioxidative Status in Epilepsy

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Abstract

Present study included 30 diagnosed patients of epilepsy and 30 normal controls in which the concentration of lipid peroxidation (malondialdehyde MDA), erythrocyte enzyme superoxide dismutase (SOD) & plasma vitamin E levels were compared. Lipid peroxidation in patients with epilepsy was significantly higher when compared with controls. Erythrocyte superoxide dismutase (SOD) and plasma vitamin E levels were significantly lower in epilepsy patient when compared with controls. This study indicates that the antioxidant status of epileptic patients was low when compared with controls.

Key words: *Epilepsy, Lipid peroxidation, Super oxide dismutase, Vitamin E.*

Introduction

An epileptic seizure can be defined as an intermittent, stereotyped, disturbance of consciousness, behavior, emotion, motor function, or sensation that on clinical ground is believed to result from cortical neuronal discharge and thus epilepsy can be defined as a condition in which seizures recur, usually spontaneously.[1]

The nervous system for a number of biochemical, physiological & anatomical reason is more vulnerable to reactive oxygen species (ROS) in addition to the other organs of the body.[2] Extensive lipid per oxidation caused due to generation of reactive oxygen species in biological membrane causes loss of fluidity, falls in membrane potential & increased permeability to H ions, leading to tissue damage.

ROS are responsible for the induction of peroxidation of unsaturated fatty acids that are components of neuronal membrane which results in depolarization.

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ROS also accelerate production of neurotoxic guanidino compounds (e.g. methylguanidine, guanidine) which are known to be convulsants in brain. Such reactions may be followed by excitatory and inhibitory neurotransmitter changes especially increased release of excitatory amino acids such as aspartic acid & decreased release of inhibitory amino acid such as GABA. These transmitter changes may be directly related to epileptogenecity by generation of ROS. These ROS in turn will accelerate production of neurotoxic guanidino compound in the pattern of a vicious cycle.[3] This shows that ROS are responsible for generation of convulsions.

Hence, the aim of the study is to evaluate the status of some antioxidants in epilepsy and the scope for antioxidant treatment.

Material & Methods

The study included 30 (19 males & 11 females) epileptic patients age and sex matched with the control group aged between 20-50 yrs from B.J. Medical College and Sassoon Hospitals, Pune.

The patients were subjected to clinical examination, Electro- encephalography (EEG) and CT scan of brain

for the diagnosis. The epileptic patients included in the present study are irrespective of their treatment status with antiepileptic drugs. Blood samples were collected in plain as well as in EDTA bulb from control and epileptic group. Blood sample in plain bulb was allowed to clot for 1 hr and then centrifuged at 2500 rpm for 10 minutes to obtain serum.

This serum was used to measure the lipid peroxidation in terms of malondialdehyde (MDA) by method of Buege et al[4] using thiobarbituric acid (TBA). Vitamin E was also determined in the serum as an antioxidant by the method of Baker & Frank et al[5] This assay is based on vitamin E extraction and reduction of ferric ions by Vitamin E.

Table 1: Comparison of lipid peroxidation and antioxidants in patients & controls.

	Normal	**Epilepsy**
Sample size	N = 30	N = 30
Serum MDA (nm/m1)	2.58 + 0.62	4.6 +0.80 **
Serum Vit. E (mg %)	1.11+0.15	0.95 +0.18*
Erythrocyte SOD (Units /gm of Hb)	**650.42+61.93**	**456+47.47****

* P < 0.01 Significant ** P < 0.001 Highly significant

Blood sample collected in EDTA bulb was centrifuged for plasma separation at 2500 rpm for 10 min. Buffy coat was removed and erythrocytes were washed thrice with normal saline. The upper layer was removed with each wash and the erythrocytes at the bottom were lysed with equal volume of distilled water. This prepared hemolysate was used for measuring Super oxide dismutase (SOD) by Marklund & Marklund method based on inhibition of auto-oxidation of pyrogallol by SOD.[6]

Data were analyzed statistically by Anova test (for age) Chi square test (for sex), unpaired T test for all parameters. For age & sex, differences of $P < 0.05$ were considered significant. For unpaired T test, differences < 0.01 were significant while $P < 0.001$ were highly significant.

Results

The control and the epileptic groups compared for age and sex were statistically insignificant. Serum malondialdehyde (MDA) levels were significantly high in the epileptic group when compared with the control group. Serum vitamin E levels were lowered in epileptic patients and the decrease was statistically significant. Erythrocyte superoxide dismutase (SOD) levels were also lowered significantly in epileptic patients compared to the control group. The decrease was statistically very significant.

Discussion

Lipid peroxidation in the neuronal membranes takes part in the mechanism of epileptic activity development has been reported by Nikushkin Braslavskii et al.[7] Tissues of the CNS may be more vulnerable to oxidative stress because of their constant high rate of oxygen consumption and high mitochondrial density. Free radicals are the byproducts of energy metabolism and the energy metabolism in brain is high. It is one of the most metabolically active organs of the body with an oxygen consumption of 35 ml/min/kg.

Brain has relatively high levels of unsaturated fatty acids that are particularly good substrates for peroxidation reactions while endogenous defense mechanisms are poor in brain. Free radicals in addition to contributing to the neuronal injury in cerebral ischemia and hemorrhage may be involved in neuronal degenerations. In epileptic seizures, excitatory amino acid receptor activation by glutamate or N-methyl, D- aspartic acid (NMDA) has been known to accompany generation of reactive oxygen species.[3]

In the present study, lipid peroxidation measured in terms of serum MDA was found to be increased significantly compared with the control group. This result is comparable with the studies done by V. Th Ramaekes et al.[8] Though the patients are on anticonvulsant therapy, the lipid peroxidation level was found to be more in the epileptic group which is in agreement with the results shown by Liu CS et al.[9] and Oztas B et al.[10]

The antioxidant Vitamin E acts as a chain breaking antioxidant. This has been reported to delay significantly

the onset of electroencephalographic seizures induced by intracerebral Ferrous chloride injection.[3] More research on vitamin E revealed that addition of vitamin E to the treatment decrease the frequency of seizure activity and also normalize the electroencephalograph (EEG). This is reported by Kovalenko VM et al.[11] In the present study, vitamin E levels were found to be decreased in epileptic patients' significantly. This result potentiates the need of addition of Vitamin E in treatment of epilepsy.

Furthermore, the other antioxidant enzyme erythrocytic superoxide dismutase (SOD) is also lowered in epileptic patients.[12] In present study SOD activity is decreased up to 31% compared to controls. The results of present study are comparable with the previous studies.

Conclusion

The epileptic activity can disturb the antioxidant status in patients irrespective of their treatment status, suggesting addition of antioxidants in the treatment.

Furthermore, the effect of anticonvulsive treatment in antioxidant level should be studied in details. The addition of antioxidants to the treatment promises the tendency of returning antioxidant balance to normal status and also decreasing the epileptic activity.

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