ROLE OF ALPHA LIPOIC ACID IN AMELIORATING OXIDATIVE STRESS IN AMITRIPTYLINE TOXICITY

1Dr.R.Vanaja, 2Dr.H.Khadar Ali

1Associate Professor, Department of Biochemistry, A.C.S. Medical College & Hospital Chennai-77.
2Research Assistant (Rtd) Institute of Pharmacology, Madras Medical College, Chennai-3

Corresponding author: E.Mail Id. drvanajaa@gmail.com Mobile No: 9840670646.

ABSTRACT

Objective: To study the effect of Alpha lipoic acid (ALA) in reducing the oxidative stress induced by amitriptyline (AP) toxicity. Methods: 100 patients of both sex & age group 25 to 60 years were selected from IMCU and toxicology wards of Government General Hospital, who were suspected to have amitriptyline toxicity. I- Group of 25 healthy volunteers of age and sex matched formed the control. II – Group of 25 patients received routine treatment (RT). III Group - RT + Vitamin C. IV Group - RT+ ALA. V Group-RT+ Vitamin C + ALA. Parameters studied were plasma cholinesterase (PchE,) Malondialdehyde (MDA), Superoxide dismutase (SOD), catalase (CAT), Glutathione peroxidase (GPX), Glutathione reductase. Results: The levels of the above enzymes and MDA showed marked alterations on treatment with ALA and Vitamin C along with routine treatment when compared with routine treatment alone. Statistical analysis of the results were carried out using Student’s ‘t’ test. Conclusion: It may be concluded that the oxidative stress induced by amitriptyline toxicity is ameliorated by supplementing with ALA and Vitamin C along with routine treatment.

Key words: Alpha lipoic acid, amitriptyline, oxidative stress.

INTRODUCTION

Acute poisoning is a common and emergency medical problem worldwide, which requires hospitalization [1]. One of the psychotropic drug is the antidepressant amitriptyline, which is a tricyclic compound. Ingestion of more than 10 mg/kg of AP produces significant toxicity. 10 – 30mg/kg is potentially lethal dose which produces sinus tachycardia, brisk reflexes, sedation, seizures, urinary retention etc [2]. Amitriptyline is used in the treatment of diabetic neuropathic pain [3]. Generally poisons produce free radicals which in turn increase oxidative stress. To counteract the effects of free radicals, antioxidants are supplemented with the routine treatment to augment the effect of the drugs. ALA is an organosulphur compound derived from octanoic acid and is essential for aerobic metabolism. It is used as dietary supplement in some countries and pharmaceutical drug in other countries. ALA is an antioxidant medication also. Several short term trials showed that it was helpful in relieving pain due to diabetic neuropathy [3]. It is considered as a potent antioxidant utilized in prevention and cure of various neurological problems.[4] It is also a free radical scavenger and possesses the ability to cross the blood brain barrier. ALA heals free radical induced mitochondrial damage. [5] It can work throughout the body because it is considered as a universal antioxidant. It is both hydrophilic and hydrophobic in nature [6]. Several lipophilic substances are used in the treatment of AP toxicity. Vitamin C exerts the direct antioxidant effect and it acts as a substrate for the enzyme ascorbate peroxidase seen in plants [7].

Considering the above beneficial effects of Vitamin C and ALA it was proposed to supplement both antioxidants along with RT to acute amitriptyline toxic patients and assess the levels of the antioxidant enzymes at the time of admission and after the treatment.
METHODS

100 patients of age 25-60 years and of both sex were chosen for this study from IMCU and Toxicology wards of Government general Hospital Chennai. Patients and their attendants gave their consent. Ethical committee approval was obtained from the institute. The patients were divided into 4 groups of 25 each. I group of 25 healthy volunteers of the same age group and sex was treated as controls.

II group of 25 amitriptyline toxic patients received RT of NaHCO₃ infusion. Activated charcoal was also infused. RT includes an alpha blocker Tamsulosin 0.4mg/day for two weeks.

III group 25 patients were treated with RT along with vitamin C 500 mg once a day for 30 days.

IV group 25 patients were given RT with 600 mg ALA once a day for 30 days.

V group 25 patients were treated with RT along with 500 mg vitamin C, and 600 mg ALA for 30 days.

All the above patients were subjected to stomach wash on arrival to the hospital and the gastric contents were analysed by thin layer chromatography to detect the presence of amitriptyline. The samples were run in parallel with the standard. The Rf values of the solute front and the solvent front of the patients were compared with those of the standard and confirmed the presence of the drug. Blood samples were collected from all the groups of controls and patients and estimated plasma cholinesterase (PchE) [8], MDA [9], CAT [10], SOD[11], GPX [12], Glutathione reductase [13], before and after the respective treatments. The chemicals and the kits were procured from Sigma Aldrich chemical Co & Hi-media Lab (Nasik – India). All the values were analysed statistically using Student’s ‘t’ test

RESULTS AND DISCUSSION

The results of the present study are elaborated in Tables 1, & 2, Table 1 explains the levels of PchE and MDA. It was noticed that there was a significant reduction (p<0.001) in the levels of PchE activity in amitriptyline toxic patients when compared with the controls. It was reported that amitriptyline inhibit PchE activity. Muscle weakness, convulsions and respiratory distress were noticed in these patients [14]. However, with RT the levels improved to a slight extent, whereas the levels showed significant improvement with vitamin C & ALA when compared against the II group of patients (p<0.001). The V group of patients exhibited near normal levels. Table I also depicts the levels of MDA which is the marker for lipid peroxidation. There was a significant increase (p<0.01) in the MDA levels of amitriptyline toxic patients when compared with the controls, indicating that the toxicity of the drug produces free radicals. The levels of MDA showed a descending trend with RT, further decline was noticed with vitamin C and ALA (p<0.01) when compared against the II group of patients. The levels were reverted to normal when RT was combined with vitamin C and ALA. Table 2 shows the levels of CAT, SOD, in controls and in patients with amitriptyline overdose, and after treating with RT, with RT & vitamin C, with RT and ALA, with RT along with vitamin C & ALA. The results showed that with amitriptyline there was a significant (p<0.01) reduction in the levels of CAT & SOD. After RT the levels did not show significant improvement (p>0.05). However, RT with vitamin C and RT with ALA showed an increase in the above levels. There was a statistically significant (p<0.01) variation noticed with RT given along with vitamin C and ALA as compared against the II group of patients.

Table 1: Levels of PchE & MDA in amitriptyline toxicity and Effect of Vitamin C and ALA.

<table>
<thead>
<tr>
<th>Group</th>
<th>PchE(U/L)</th>
<th>MDA(n moles/ml)</th>
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<tbody>
<tr>
<td>Control</td>
<td>5600±500</td>
<td>180±23.21</td>
</tr>
<tr>
<td>Amitriptyline</td>
<td>1010±150***</td>
<td>260±13.11**</td>
</tr>
<tr>
<td>RT</td>
<td>1800±78**</td>
<td>212±11.23*</td>
</tr>
<tr>
<td>RT+Vitamin C</td>
<td>2300±60**</td>
<td>203±10.4*</td>
</tr>
<tr>
<td>RT + ALA</td>
<td>3850±58**</td>
<td>192±9.8*</td>
</tr>
<tr>
<td>RT+vitamin C+ALA</td>
<td>5100±100***</td>
<td>184±9.4**</td>
</tr>
</tbody>
</table>

Values are Mean ± SD *----- P< 0.05-significant **-----P<0.01-significant ***----P<0.001-highly significant
Table 2 also explains the levels of GPX, & Glutathione reductase, in the control, amitriptyline toxicity, treatment with routine drugs, RT with vitamin C, RT with ALA and RT given along with vitamin C and ALA. The results showed similarity with those seen with CAT & SOD, as explained in Table 1. The mechanism of action of vitamin C in promoting the antioxidant status may be due to the chain propagation activity and rapid reaction with the oxygen free radicals formed due to the excessive dose of the drug. Vitamin C scavenges the free radicals and inhibit lipid peroxidation. Amitriptyline provokes increase of intracellular lipid peroxidation [15]. Recently it was shown that amitriptyline toxicity is through mitochondrial dysfunction and ROS production [16]. Generally the increase in ROS brings about increase in antioxidants. But due to the high rate of ROS input, it leads to the reduction of antioxidant enzymes. Hence very low levels of CAT, SOD, GPX, Glutathione reductase, were produced in amitriptyline toxicity [17], because the poison provokes apoptosis and significant reduction in antioxidant level [18]. However when ALA was also supplemented along with vitamin C, the status of the antioxidants was better which may be due to the fact that ALA in addition to its own antioxidant properties, also recycles vitamin C & Vitamin E thereby enhancing their availability both in aqueous phase (cytosol) and lipid phase (cell membrane).

<table>
<thead>
<tr>
<th>Group</th>
<th>CAT (µ moles /mg Hb)</th>
<th>SOD (µ moles /mg Hb)</th>
<th>GPX (µg/ml)</th>
<th>Glutathione reductase (µg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>4.28±0.1</td>
<td>6.5±0.74</td>
<td>6.43±0.72</td>
<td>9.58±0.57</td>
</tr>
<tr>
<td>Amitriptyline</td>
<td>3.0±0.08*</td>
<td>4.2±0.32**</td>
<td>4.8±0.23**</td>
<td>7.4±0.42**</td>
</tr>
<tr>
<td>RT</td>
<td>3.62±0.55Ns</td>
<td>4.5±0.84Ns</td>
<td>5.2±0.21Ns</td>
<td>8.0±0.21Ns</td>
</tr>
<tr>
<td>RT+Vitamin C</td>
<td>4.0±0.23*</td>
<td>5.0±0.5*</td>
<td>5.6±0.14*</td>
<td>8.4±0.23*</td>
</tr>
<tr>
<td>RT + ALA</td>
<td>4.2±0.31*</td>
<td>5.8±0.58*</td>
<td>6.0±0.22**</td>
<td>9.9±0.30**</td>
</tr>
<tr>
<td>RT+vitamin C+ALA</td>
<td>4.4±0.73**</td>
<td>6.4±0.22**</td>
<td>6.6±0.23**</td>
<td>10.12±0.40**</td>
</tr>
</tbody>
</table>

Values are Mean ± SD *---- P< 0.05-significant **-----P<0.01-significant ***----P<0.001-highly significant Ns-not significant

SUMMARY AND CONCLUSION

The present study revealed that in amitriptyline toxicity, the supplementation with vitamin C and ALA along with the RT improved the antioxidant status of the patients when given additively than when given individually.

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REFERENCES


