Phytochemical, Antioxidant, Anti-stress and Cerebro-protective activity of ethyl acetate extract of bark of *Entada rheedi*

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**ABSTRACT**

*Entada rheedi* is claimed to have antistress activity by folklore which is available abundantly in several places of India. The present study was planned to evaluate the phytochemical, anti-oxidant, anti-stress and cerebroprotective activities of ethyl acetate extract of bark of *Entada rheedi* (EAER). The bark of *Entada rheedi* was collected and extracted with ethyl acetate. The ethyl acetate extract was subjected to phytochemical screening (chemical and HPTLC), antioxidant (*in-vitro*), anti-stress (mice model) and cerebroprotective activities (cerebral ischemia model). EAER showed the presence of flavonoids as primary phytoconstituents. EAER significantly reduced the immobility time in swimming endurance and tail suspension test. EAER significantly reduced the TBARS levels and augmented tissue antioxidants in restraint stress model and cerebral ischemia model. The levels of MOA-A were reduced in the EAER treated animals and cortisol levels also reduced in EAER treated animals. Histopathology also supported the biochemical parameters. The EAER effect was compared with reference standard diazepam and Ashwagandha. EAER showed significant antioxidant, anti-stress and cerebroprotective activities and the protective effect could be due to the presence of flavonoids as phytoconstituents.

**INTRODUCTION**

Stress is a state or feeling practiced by a person when he observes the demands that exceed the personal and social possessions that the individual is capable of mobilizing. Stress distracts the normal physiological state and results in a condition of susceptible homeostasis (Habbu et al., 2010). Stress enhances the action of the endocrine system to release a high amount of glucocorticoids and catecholamines. The continuous state of stress leads to cause abnormal physiological state development of psychological and cognitive dysfunction (Nade et al., 2009; Singh et al., 2009; Joshi et al., 2012). Furthermore, during stressful situations, the energy necessities of the body are augmented, leading to improved production of reactive oxygen species (ROS) in the body resulting in the progress of oxidative stress. These ROS can cause tissue injury by reacting with proteins, lipids, DNA resulting in various pathological situations (Koppula et al., 2009; Joshi et al., 2012). Numerous measures are existing to counteract the possessions of stress, which comprise non-pharmacological and pharmacological methods (Nade et al., 2009). Use of some anti-stress agents, despite presenting significant anti-stress activity beside various models of stress, have not showed effective in contradiction of chronic stress-induced side effects on male sexual function, immunity, during pregnancy and lactation, behavior...
cognition (Habbu et al., 2009; Nade et al., 2009; Desai et al., 2011). Consequently, there is a necessity for an active herbal anti-stress drug in the treatment of stress-induced illnesses (Habbu et al., 2009). The possible usefulness of harmless and inexpensive herbal drugs has been reported for anti-stress activity. Various herbs such as Asparagus racemosus, Ocimum sanctum, Piper longum, Tribulus Terrestris, Withania somnifera were claimed to have anti-stress activity with several pharmacological actions (Desai et al., 2011).

Entada rheedi Spreng (Fam. Mimosaceae) is a woody climbing shrub that grows naturally in Africa, tropical Asia, Australia and a small part of the Pacific islands (Ghani 1998; Uddin 2006). It has been reported useful in the treatment of jaundice, diarrhea, musculoskeletal problems and mumps. It is also used as a remedy for cerebral hemorrhage and oral contraceptive (Nzowa et al., 2010). The bark of Entada rheedi contains saponin, which is used as a substitute for soap (Ahmed, 2009).

The present research was intended to assess the anti-oxidant, anti-stress and cerebroprotective activities of ethyl acetate extract of bark of Entada rheedi in experimental animal models.

MATERIALS AND METHODS

Animals

Male Wistar albino rats (200±10 g) and male Swiss albino mice (25 ± 5 g) were used for the research. All the animals were administered with standard laboratory food and water ad libitum (Sai Durga Feeds and Foods, Bangalore). The animal experimentation was carried out with the prior approval from the Institutional Animal Ethics Committee with the approval number of 1220/a/08/CPCSEA/ANCP-08/15.

Plant material and preparation of extract

The bark of Entada rheedi was obtained from the forest area of Tirumala in the month of Feb 2016 and was validated by Dr. K. Madhava Chetty, Professor and Head, Department of Botany, S. V. University, Tirupati. The sample of plant material was deposited in the department with the number (Plant specimen number: 1101) for future reference. 500 g of shade-dried course powder of the bark of Entada rheedi was extracted with various solvents like petroleum ether and ethyl acetate by soxhalation method. The yield of the ethyl acetate extract of Entada rheedi (EAER) was found to be 10.8%.

The preliminary phytochemical screening of EAER was performed by the methods described by Harbone & Baxter, 1993 for various phytoconstituents. Sample (5 μL-10 mg/5 ml) of EAER was analyzed to identify the specific constituents by the method HPTLC by using chloroform: methanol (6:4) as mobile phase. The established plate was parched in the oven at 60°C to vanish solvents from the plate. The plate was reserved in the photo-documentation chamber (CAMAG REPROSTAR 3), and pictures were taken under ultraviolet (UV) light at 254 and 366 nm, correspondingly. The Rf values and fingerprint data were logged by WIN CATS software.

Estimation of Flavonoids

The experiment was performed to identify the total flavonoid content by using rutin as standard flavonoids by the method described by (Helmja et al., 2007).

In vitro antioxidant activity of EAER

The reducing power of EAER was studied by adopting the method of Oyaizu 1986. The free radical scavenging activity of EAER was measured in terms of hydrogen donating or radical scavenging ability using the stable radical DPPH (Burits & Bucar, 2000). The peroxide radical scavenging activity of EAER was carried out by adapting the method of Ruch et al., 1989. The nitric oxide scavenging activity was estimated by the method described by (Marcocci et al., 1994). Superoxide radical scavenging activity was determined by the NBT reduction method (Misra& Fridovich, 1972).

The percentage inhibition was calculated from the following equation;

\[
\% \text{ inhibition} = \frac{A_{\text{blank}} - (A_{\text{sample}}/A_{\text{blank}})}{A_{\text{blank}}} \times 100
\]

Acute oral toxicity study

The LD50 value of EAER was determined by the method described by Lorke, 1983. EAER up to a dose of 2000 mg/kg did not show any signs of deadliness and death.

Anti-stress activity

Chemical-induced stress in mice

The antistress activity was assessed for EAER by using chemical induced, swimming endurance induced stress (Nimbkar et al., 2001) and tail suspension test (Amogh et al., 2016) in mice. The mice were randomly divided into four groups of six animals each. Group I received 2% tween 80 for 7-days and served as vehicle control. Groups II and III received different doses (200, and 400 mg/kg p.o) of EAER for 7-days. Group IV received diazepam (2 mg/kg i.p) for 7-days and served as a
positive control. On day 7, 1 h after the drug treatment, the antistress activity was determined by reduction in writhing in chemical induced stress and reduction in immobility time in swimming endurance test and tail suspension test.

**Restraint stress in mice**

In the restraint stress model, the mice were treated with EAER for 15 days. On the 15th day, 1 h after the last treatment, the forelimbs and hind limbs of mice in all groups except control group were tied using adhesive tape for 2 h to induce stress by immobilizing them (Bhattacharya et al., 1999). The adhesive tapes were removed after 2 h and blood was collected from retro-orbital plexus of all the animals to evaluate biochemical parameters. The mice were then sacrificed under ether anaesthesia and their brain, adrenal glands and spleen were isolated and weighed.

**Cerebral ischemia model**

**Experimental Protocol**

The animals were divided into 4 groups (n=6 per group). Group –I received 2% tween 80 (10 ml/kg p.o) and served as normal control, Group-II received iron and served as disease control, Group-III received EAER 400 mg/kg p.o. Group–IV received the *Withania Somnifera* 100 mg/kg p.o for 28 days. Iron dextrose injection (0.5 mg/kg i.p) was administered 24 hrs prior to the carotid artery occlusion, then the animals were subjected for carotid artery occlusion, for 15 min followed by reperfusion 30 min prior to scarification. Then the animals were sacrificed, and the parameters were studied (Traystman, 2003).

**Assessment of biochemical parameters**

The blood samples were centrifuged (3000 rpm for 20 min) and the serum obtained was separated and used for the estimation of blood glucose, total cholesterol, and triglycerides using commercial kits.

**Assessment of tissue antioxidant parameters**

The isolated mice and rat brains were rinsed with 0.9% ice-cold normal saline and processed to get 10% homogenate in cold phosphate buffer using glass Teflon homogenizer. The homogenates obtained were used for the estimation of TBA reactive substance (TBARS) (Okhawa et al., 1979), reduced glutathione (GSH) (Ellman, 1959), superoxide dismutase (SOD) (Kakkar et al., 1984), catalase (Aebi, 1974), protein (Bradford, 1976), and nitrate (Green et al., 1982). Estimation of Monoamine Oxidase A was performed by the method described by Habibur & Ewaraiah, 2008. Serum corticosterone(CORT) level was determined quantitatively using rat ELISA kits (Dhingra et al., 2014).

**Measurement of the infarcted area**

After sacrafication rat brains were isolated and washed with ice-cold saline to remove blood and adherent tissue. 2% solution of 2,3,5-triphenyl tetrazolium chloride (TTC) was prepared using phosphate buffer (PH 7.4). The brains were then kept in TTC solution overnight. Infract sizes caused by cerebral ischemia were visualized by TTC staining. After 24 h brains were taken out from the dye and the infarcted area was observed by naked eye where the normal cells will receive stain and visualised in red colour while the infarcted cells will not receive the stain and visualized in a pale colour.

**Histopathology**

At the end of the study, the brain tissue was fixed in 10% formalin and embedded in paraffin wax and sent for histopathological studies.

**Statistical analysis**

Results were expressed as mean ± standard deviation and analyzed using Graph Pad Prism version 5.1 Graph Pad Software, Inc using one-way analysis of variance followed by Dunnett’s posttest. *P* < 0.05 was considered to be significant.

**RESULTS**

**Preliminary phytochemical screening**

The preliminary phytochemical analysis revealed the presence of alkaloids, carbohydrates, saponins, phenolic compounds, and flavonoids.

**High-performance thin layer chromatography**

**Fingerprint analysis of EAER**

![Figure 1: HPTLC fingerprint analysis of EAER at 366 and 254 nm](image)

High-performance thin layer chromatography fingerprinting analysis of EAER revealed several peaks and were recorded. HPTLC profile under UV
The Rf value of spots observed were 0.13, 0.80 and 0.92. The appearance of blue and orange color under UV examination confirmed the presence of flavone and flavonoid components in the extract.

### Flavonoid content of EAER

Flavonoid content of EAER was determined, and the results indicated that EAER contains 18.0 mg/g of rutin equivalent flavonoid.

The results of in-vitro antioxidant activity of EAER are presented in Table 1.

| Table 1: In-vitro antioxidant activity of EAER
<table>
<thead>
<tr>
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<tbody>
<tr>
<td>Concentration (µg/mL)</td>
<td>Reducing Power (OD)</td>
<td>DPPH assay (%)</td>
<td>Superoxide radical scavenging assay (%)</td>
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<tr>
<td>-------------------</td>
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<td>-----------------</td>
<td>-----------------</td>
</tr>
<tr>
<td>EAER</td>
<td>AA</td>
<td>EAER</td>
<td>AA</td>
</tr>
<tr>
<td>10</td>
<td>0.14</td>
<td>0.234</td>
<td>40.31</td>
</tr>
<tr>
<td>50</td>
<td>0.168</td>
<td>0.647</td>
<td>41.77</td>
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<td>0.19</td>
<td>0.782</td>
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<td>0.196</td>
<td>0.675</td>
<td>53.18</td>
</tr>
<tr>
<td>500</td>
<td>0.21*</td>
<td>2.826</td>
<td>75.35*</td>
</tr>
<tr>
<td>IC50</td>
<td>---</td>
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<td>85</td>
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</table>

EAER - ethyl acetate extract of Entada rheedi; AA- Ascorbic acid

| Table 1: In-vitro antioxidant activity of EAER (Contd..)
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<tr>
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<tr>
<td>Concentration (µg/mL)</td>
<td>H2O2 radical scavenging assay (%)</td>
<td>Nitric oxide inhibition assay (%)</td>
<td>Hydroxyl radical scavenging assay (%)</td>
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<tr>
<td>EAER</td>
<td>AA</td>
<td>EAER</td>
<td>AA</td>
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<td>10</td>
<td>86.68</td>
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<tr>
<td>50</td>
<td>88.33</td>
<td>33.33</td>
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<tr>
<td>100</td>
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<td>41.98</td>
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<td>94.17</td>
<td>50</td>
<td>77.26</td>
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<tr>
<td>500</td>
<td>97.5*</td>
<td>58.33</td>
<td>89.28*</td>
</tr>
<tr>
<td>IC50</td>
<td>6200</td>
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<td>125</td>
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</tbody>
</table>

EAER - ethyl acetate extract of Entada rheedi; AA- Ascorbic acid

| Table 2: Effect of EAER on different biochemical parameters in brain homogenate
<table>
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<tbody>
<tr>
<td>Treatment group</td>
<td>TBARS (nmol/g wet Wt)</td>
<td>NO (µg/dl)</td>
<td>Protein (mg/dl)</td>
<td>GSH (µg/g wet Wt)</td>
<td>SOD (IU/mg Protein)</td>
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</tr>
<tr>
<td>Vehicle control</td>
<td>48±2.94</td>
<td>32±0.42</td>
<td>96±3.69</td>
<td>236±9.2</td>
<td>14.6±1.02</td>
</tr>
<tr>
<td>Stress control</td>
<td>126±9.4a</td>
<td>72±1.08a</td>
<td>75±1.38a</td>
<td>93±1.7a</td>
<td>5.7±0.4a</td>
</tr>
<tr>
<td>EAER 200 mg/kg</td>
<td>86±1.62b</td>
<td>42±1.61b</td>
<td>82±1.14b</td>
<td>179±5.7b</td>
<td>10.8±0.7b</td>
</tr>
<tr>
<td>EAER 400 mg/kg</td>
<td>52±0.86b</td>
<td>26±0.74b</td>
<td>96±1.49b</td>
<td>214±10.5b</td>
<td>16.8±1.02b</td>
</tr>
<tr>
<td>Diazepam 2 mg/kg</td>
<td>61±0.62bc</td>
<td>30±1.02bc</td>
<td>88±0.82bc</td>
<td>169±1.8bc</td>
<td>12.1±1.08bc</td>
</tr>
</tbody>
</table>

All values expressed as mean±SEM. One-way ANOVA followed by Dunnet’s post-test. a P<0.001 vs Vehicle control; b P<0.001 vs stress control

| Table 3: Tissue level of Catalase, GSH, SOD, TBARS and Nitrates
<table>
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<tbody>
<tr>
<td>Group</td>
<td>TBARS (nmol/g wet Wt)</td>
<td>GSH (µg/g wet Wt)</td>
<td>SOD (IU/mg Protein)</td>
<td>Catalase (IU/mg protein)</td>
<td>NO (µg/dl)</td>
</tr>
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<td>-----------------</td>
<td>-----------------</td>
<td>-----------------</td>
<td>-----------------</td>
<td>-----------------</td>
<td>-----------------</td>
</tr>
<tr>
<td>Vehicle control</td>
<td>24.15±2.138</td>
<td>34.46±1.11</td>
<td>21.52±2.387</td>
<td>0.120±0.177</td>
<td>0.396±0.014</td>
</tr>
<tr>
<td>Stress control</td>
<td>33.08±2.278a</td>
<td>18.78±0.42a</td>
<td>9.520±0.144a</td>
<td>0.024±0.013a</td>
<td>0.746±0.024a</td>
</tr>
<tr>
<td>EAER 400 mg/kg</td>
<td>20.020.06bc</td>
<td>22.43±1.063d</td>
<td>11.91±0.721c</td>
<td>0.133±0.001c</td>
<td>0.471±0.022d</td>
</tr>
<tr>
<td>WS 100mg/kg</td>
<td>19.29±0.936c</td>
<td>23.25±0.9361b</td>
<td>18.713±1.644c</td>
<td>0.162±0.007c</td>
<td>0.495±0.019c</td>
</tr>
</tbody>
</table>

All values expressed as mean±SEM. One-way ANOVA followed by Dunnet’s post-test. a P<0.001 vs Control; b P<0.05 vs Disease control; c P<0.01 vs Disease control; d P<0.001 vs Disease control

366 and 254 nm was recorded in Figure 1 and 2. The Rf value of spots observed were 0.13, 0.80 and 0.92. The appearance of blue and orange color under UV examination confirmed the presence of flavone and flavonoid components in the extract.
Figure 2: HPTLC fingerprint analysis of EAER

**In-vitro** antioxidant activity of EAER

Figure 3: Effect of EAER on chemical induced stress; (All values expressed as mean±SEM. One-way ANOVA followed by Dunnet’s post-test. **P<0.001 vs Stress control)

Anti-stress activity

**Chemical-induced stress:** Oral administration of EAER in two different doses significantly (P<0.001) reduced the writhes induced by acetic acid in mice. The percentage inhibition at the high dose of EAER (77.34%) was incomparable to reference standard diazepam (85.28%). The results are presented in Figure 3.

**Swimming endurance test:** Oral administration of EAER in two different doses significantly (P<0.001) reduced the immobility time in mice during the swimming endurance test. The percentage inhibition at the high dose of EAER (67.81%) was incomparable to reference standard diazepam (61.64%). The results are presented in Figure 4.

**Tail suspension test:** In tail suspension test, seven-days pre-treatment with two doses (200 mg/kg, 400 mg/kg) of EAER significantly reduced the immobility time as compared to the stress control group. *Withania somnifera* (100mg/kg) showed a significant reduction in immobility time as compared to the stress control group. The results were shown in Figure 5.

Figure 6: Effect of EAER on Glucose, Total cholesterol & Triglycerides in the blood; (All values expressed as mean±SEM. One-way ANOVA followed by Dunnet’s post-test. a P<0.001 vs Vehicle control; b P<0.001 vs Disease control)
Restrain stress model: Oral administration of EAER in two different doses significantly reverted the biochemical parameters like glucose, cholesterol and triglyceride in blood serum near to control group. The results are presented in Figure 6.

Effect of EAER on different biochemical parameters in brain homogenate (Table 2)

There was significant (P<0.001) increase in the level of TBARS and NO in brain homogenate of stress group. The animals treated with EAER in two different doses (200 and 400 mg/kg BW) significantly (P<0.001) reduced the level of TBARS and NO, and the results are similar to the reference standard diazepam.

There was significant (P<0.001) decrease in the level of protein, GSH, SOD and catalase in brain homogenate of stress group. The animals treated with EAER in two different doses (200 and 400 mg/kg BW) significantly (P<0.001) augmented the level of protein, GSH, SOD and catalase in brain homogenate and the results are similar to the reference standard diazepam.

Results of tissue antioxidant parameters in cerebral ischemia model (Table 3).

Level of Thiobarbituric acid reactive substances (TBARS)

There was a significant (p<0.001) increase in the level of TBARS in disease control when compared with normal control. There was less significant (p<0.001) increase in the level of TBARS in groups treated with EAER and standard drug.

Level of reduced glutathione: There was a significant (p<0.001) decrease in the level of GSH in disease control when compared with normal control. There was less significant (p<0.001) increase in the level of GSH in groups treated with EAER and standard drug when compared with disease control.

Level of superoxide dismutase: There was a significant (P<0.001) decrease in the level of the enzyme catalase in disease control when compared with normal control. There was a less significant (P<0.001) increase in the level of catalase in groups treated with EAER and standard drug.

Nitrates level: There was a significant (p<0.001) increase in the level of nitrates in disease control when compared to normal control. There was significant (P<0.001, P<0.01) decrease in the level of nitrates in the group treated with EAER and standard drug.

Level of monoamine oxidase A in brain tissue (Table 4)

There was a significant (p<0.001) increase in the level of MAO-A between disease control when compared with normal control. There was less significant (p<0.001) decrease in the level of MAO-A in groups treated with plant extract and standard drug.

Level of CORT in serum (Table 4): There was a significant (p<0.001) increase in the level of CORT between disease control when compared with normal control. There was less significant (p<0.001) decrease in the level of CORT in the group treated with plant extract and standard drug.

Measurement of Infarcted Area

Figure 7: Results of infarct size area of the brain

The brain tissue showed the less infarcted area in EAER (C), and WS (D) when compared with the disease control (B) which contains unstained infarcted cells. The results were presented in Figure 7.

Figure 8: Histopathological results of brain tissue
Histopathological Evaluation

Photomicrograph of rat brain showed normal architecture with regular morphology of brain cell membrane and well-preserved cytoplasm in control group (A). The disease control group (B) showing disruption of epidermal cell integrity, and damage to the neuronal cells and glial cells. Rats pre-treated with EAER (C), WS (D) have shown no damage to the neuronal and glial cells with normal epidermal cell integrity similar to that of the normal control group. The results were presented in Figure 8.

DISCUSSION

Stress is a major problem globally which arise through several factored to affect human health and organ system (Debnath et al., 2011). The short duration of stress does not cause any harm to body but longer duration of stress cause development of abnormal conditions like anxiety, behavioural depression, cognitive dysfunction, and increase in serum corticosterone level, increase in blood glucose, immune suppression, gastric ulcers and increased oxidative stress (Debnath et al., 2011). Many medicinal plants show effectiveness against the developed stress which produced harm condition to the body (Anju, 2011).

Entada rheedi is claimed to have antistress activity by folklore which is available abundantly in several places of India. Powdered seeds of Entada rheedi mixed with water is given for puerperal fevers. Several medicinal properties were reported with the plant like skeletal muscle problem, liver diseases and dental problem as an ointment.

In the present study, the anti-stress activity of EAER (200 and 400 mg/kg) has been assessed by using experimental animals. The screening tests were used in the present study was physical and restraint, screening models. Acetic acid induced writhing test and swimming endurance test are the established physical stress models for the assessment of anti-stress activity.

Mice are used as an experimental animal model for physically induced stress. Animals were imposed for forced swimming to identify the various behavior like immobility which related to swimming stress-induced changes in behavior. The time of immobility was noted, and the drug effect was assessed as reduction in immobility time (Patel et al., 2011). In the present study have shown that there was an upsurge in swimming endurance which specifies clearly that both doses of the EAER have an anti-stress effect. The extract shows dose-dependent anti-stress activity.

Development of hyperalgesia is one of the indications of stress by acetic acid administration to the animals. The characteristic writhing was identified as hyperalgesia and animals shows the number of writing was reduced by administration of extracts. A similar finding was observed in the present study that EAER significantly reduced the number of writing produced by acetic acid with mice. A number of medicinal plants have been reported with anti-stress activity related to this experimental model (Azmathulla et al., 2006).

Stress also affects the biochemical parameters like blood glucose and triglycerides by the various mechanism. Majorly the abnormal level of blood glucose and triglycerides were due to adrenal gland activity by increasing the activity of cortisol in the blood. The adrenal gland also increases the level of catecholamines consider as stress hormones which affect the concentration of blood sugar, cholesterol and triglycerides by gluconeogenesis and lipogenesis mechanism (Anju, 2011).

Increase in the level of blood glucose, cholesterol and triglycerides were observed in the present study due to restraint stress. A similar effect also reported by many researchers in their study (Azmathulla et al., 2006; Anju, 2011; Patel et al., 2011). Administration of EAER significantly reduced the increased level of blood parameters as evidence of anti-stress activity.

Because of low antioxidant capacity and high oxygen tension, the nervous system is damaged due to stress-induced peroxidation. Reduction of intracellular glutathione is greater by lipid peroxidation and can be linked to oxidative injury as glutathione is an endogenous non-enzymatic antioxidant. In the restraint stress model of the present study, it was found that lipid peroxidation (TBARS) and nitric oxide levels were extremely augmented in the stress control group as a result of oxidative damage. The similar results were reported by many other researchers (Azmathulla et al., 2006). EAER significantly reduced the TBARS and nitric oxide levels indicating that the extracts have anti-stress activity through reduction of oxidative stress. Restraint stress also caused the diminution of glutathione, SOD, catalase and protein content in mice brain. EAER significantly increased the reduced GSH, SOD and catalase levels.

In the present study, the cerebral ischemic control group animals showed decreased amount of antioxidant defense enzymes (SOD, Catalase, GSH) and increased amount of lipid peroxidation (TBARS) in brain because of induction of ischemia whereas the group of animals that were treated with EAER, and WS had shown increased levels of SOD, Catalase, GSH enzymes and decreased TBARS. This may be due to the antioxidant principles in brain regions that are associated with motor and sensory such as...
flavonoids and phenolic compounds present in the plant extract.

Corticosterone (CORT) is a significant stress hormone in animals and has been convoluted in major depressive disorder. CORT is an endocrinological diagnostic marker which is frequently used based on its rise in chronic stress in animal models (Xu et al., 2015). Two isoforms of monoamine oxidase (MAO-A and MAO-B) exist, with complex substrate preference of MAO-A isoform for serotonin; and it is the main target for the antidepressant MAO inhibitors. The metabolic deprivation of serotonin and catecholamines in noradrenaline and serotonin neurons is structured by MAO-A. This pathway is supposed to be the site of therapeutic exploit in stress (Habbe, 2011). In the present study, cerebral ischemic rats showed a significant increase in serum level of CORT and brain activity of MAO-A relative to control rats. This effect was nearly restored by oral administration of EAER and standard Withania somnifera (WS) relative to cerebral ischemic rats, which reflected the antistress-like potential.

In this study infarct size as measured with TTC staining was used as the primary experimental measure of the outcome of cerebral artery occlusion. EAER and WS extract significantly lowered the infarct size and the extent of edema in ischemic rats as compared to the control rats. A similar effect was observed in the histopathological report also.

The HPTLC and flavonoids estimation proved that the EAER extract enriched with flavonoid type compounds which might be responsible for the above said activities.

CONCLUSION

The present study concluded that the ethyl acetate extract of Entada rheedi possesses significant antioxidant, anti-stress and cerebroprotective activities due to the presence of flavonoids as major phyto constituents.

Acknowledgement

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Conflict of interest

The authors declare no conflict of interest.

Abbreviations used


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