ABSTRACT
Psychopharmacological drugs are gaining importance in our day to day life. Drugs from natural sources are having lesser side effects compared to synthetic drugs. Sarcostemma acidum (Roxb) Voigt is a plant used in folklore practices for psychiatric conditions. The methanol extract of the whole plant of Sarcostemma acidum (MESA) was evaluated for psychopharmacological properties. MESA (200 Mg/kg), (100 mg / Kg) Diazepam (2mg / Kg), Haloperidol (5 Mg/Kg) and CMC 1% was used for the study. Actophotometer, Elevated plus maze, Hole board test was used to test CNS inhibitory activity and anxiolytic activity. The cataleptic activity was assessed by BAR test.

It was found that the methanolic extract showed an increase in the number of entries into the open arm in Elevated plus maze, compared to control group. The number of Head pocking in Hole board apparatus increased which indicate anxiolytic activity and exploratory behaviour. Reduction in loco motor activity in Actophotometer showed CNS depressant property and increase in cataleptic effect reflects extrapyramidal like effects of the plant extract.

KEYWORDS: Sarcostemma Acidum, Methanol extract, Psychopharmacology, Elevated Plus Maze, Catalepsy.

INTRODUCTION
Sarcostemma acidum (Roxb) Voigt, a xerophytic plant belongs to Asclepidaceae family, having several medical uses. It is used as an emetic and also in snake bites. It is known as soma plant to prepare “Somaras”, a rejuvenating drink by Aryans. The score of soma plant is a mystery which has been debated by vedic people and scholars. It is found as a wild plant...
in India, Pakistan and Europe, in rocky places of Bihar, Bengal, Konkan, Decan, Tamil Nadu and Kerala. The plant is bitter, acrid, cooling, emetic, antiviral and rejuvenating. The various chemical constituents reported are Malic acid, Succinic acid, Alkaloids, Phytosterols, Amyrins, Lupeol etc.\cite{3,4}

A phytochemical study was done with the whole dried plant of Sarcostemma Acidum collected from a source near Tamil Nadu, India and was authentified by Prof. Rojimon Thomas, Department of Botany, C.M.S College, Kottayam. For determining the major class of phytochemicals of Sarcostemma acidum, standard phytochemical methods were used. Both aqueous and methanolic extracts were tested and methanolic extract was confirmed by thin layer chromatography. The preliminary screening showed the presence of carbohydrates, Triterpenoids, Phytosterols, Flavanoid glycosides, Saponins and Lignins in Sarcostemma Acidum. From the methanolic extract a flavanoid glycoside was identified. The flavanoid glycoside was isolated and further spectral characterization was done and it was concluded that the glycoside was (2R, 3R) – 3, 5, 7 Trihydroxy – 2- (3, 4, 5 trihydorxy phenyl) – 2 -3, dihydro chrome 4 – one with molecular mass 320.0529.\cite{5} Methanolic spray dried extract was used for Psychopharmacological studies using Wistar Rats. The study includes Elevated Plus Maze, Actophotometer, Hole board test and Catalepsy test.\cite{6} In earlier days Sarcostemma acidum was used by tribal people to have a sound sleep at night. The present study substantiates the traditional usage of this plant.

**MATERIALS AND METHODS**

**Animals:** Wistar Albino rats weighing 150-250 gms of either sex, maintained under standard husbandry conditions, grouped randomly were used for the screening which was obtained from animal house of University College of Pharmacy, Cheruvandoor, Kottayam, Kerala. The animals were fed with standard laboratory food during study period.

The observations were made at room temperature in a noiseless atmosphere. The study was approved by Institutional Animal Ethics Committee, University College of Pharmacy, Cheruvandoor, Kottayam, Kerala approval no: IAEC/PhD/DPS/2018-02 as per provisions of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), New Delhi, India.
Plant Materials and Extraction
Whole plant of Sarcostemma acidium (ROXB) Voigt was collected locally from Tamil Nadu, India and was authenticated by Professor Rojimon Thomas, Department of Botany, CMS College, Kottayam. The plant was air dried in shade and powdered. 10 gm of air dried powdered material was taken in a thimble and kept in soxhlet apparatus. It was consecutively extracted with 250 ml each of Petroleum Ether, Hexane, and Ethyl Acetate for 4 hours. All fractions were discarded and finally extracted with Methanol for 4 hours. The Methanol extract was concentrated and dried.[5,6,7]

Chemicals and Drugs Used: Diazepam, Carboxy Methyl Cellulose, Haloperidol.

Pharmacological evaluation: Acute oral toxicity study
Acute oral toxicity study was performed as per OECD 423 Guidelines. Wistar rats 150-250mg, n=6 of either sex was selected for the study. The animals were kept fasting overnight providing water only. 2000mg/kg dose of the extract, suspended in 1% Carboxy Methyl Cellulose was administered orally. The animals were observed for mortality and significant changes for 14 days. No mortality or significant changes occurred. The dose was fixed.

Psychopharmacological Evaluation
Anxiolytic activity – Elevated Plus Maze Test
The Elevated plus Maze Test is used for anxiety testing. Four groups of rats, each group containing 6 animals were taken. Standard 5 minute test was employed for each animal. The apparatus consisted of two open arms (35x6 cm) and two enclosed arms (35x6x15 cms), that extended from a common central platform. The floor is made of wood. The entire maze is elevated to a height of 40 cms above the floor level. On the first day the rats were allowed to explore the maze for 30 seconds and the transfer latency was recorded. Second day the rats were administered orally with CMC 1%, Diazepam 2mg/kg, MESA 200mg/kg, MESA 100mg/kg and placed on the maze (n=6). The transfer latency of each group was recorded.[8]

Hole Board Test- For Exploratory Behaviour
A locally constructed Hole Board apparatus was used. It consisted of 16 holes each of 3 cms diameter. The apparatus was placed in elevated position. The groups were administered orally with CMC 1%, Diazepam 2mg/kg, MESA 200mg/kg, MESA100mg/kg (n=6). The number of head dips were measured for 5 minutes.
Locomotor activity – Actophotometer.

Actophotometer was used to measure the spontaneous locomotor activity of the animal. The activity was measured after oral administration of CMC 1%, Diazepam 2mg/kg, MESA 200mg/kg, MESA 100mg/kg to each group (n=6). The units of activity was based on the number of cut off of light beam, when the animal was placed in chamber. The time of measurement was 10 minutes.

Catalepsy test – Antipsychotic activity

The catatonic activity, the failure to correct an unusual posture was measured by Bar Test. Four groups of rats, each group containing 6 animals were taken. Standard 5 minute test was employed for each animal. The groups each of 6 animals were administered orally with CMC 1%, Haloperidol 5mg/kg, MESA 200mg/kg, MESA100mg/kg. The catatonic activity was measured by placing the forelimbs of the animal on a bar elevated to 9 cm. The length of time the animal maintained this posture was recorded at 0, 30, 60, 90, 120 minutes.

Data analysis: The data was analysed using one way Anova and the groups were compared with control by using Dunnetts Post hoc test. P value of at least 0.05 was considered as level of significance in all analysis.

RESULTS

Elevated plus maze model

The methanolic extract of Sarcostemma Acidum (Roxb.) Voigt, MESA200mg/kg and MESA 100mg/kg and standard drug Diazepam 2mg/kg significantly increased the number of entries into the open arm and time spent in the open arm of the elevated plus maze, compared to the control group. The standard drug showed maximum activity, followed by MESA 200 and MESA100. The results are shown in the table. The test group MESA100, showed least number of entries into open arm and time spent in open arm compared to control group which received 1% CMC.

The methanolic extract of Sarcostemma Acidum exhibited antianxiety activity in both high and low doses in Wistar Albino rats.
Table 1. Elevated plus maze test for Anxiolytic Activity.

<table>
<thead>
<tr>
<th>Group</th>
<th>Mean of entries in</th>
<th>Mean time spent in (sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Open arm</td>
<td>Closed arm</td>
</tr>
<tr>
<td>Control</td>
<td>1.833±0.4014</td>
<td>2.500±0.2236</td>
</tr>
<tr>
<td>Diazepam2mg/kg P.O</td>
<td>5.000±0.4472**</td>
<td>1.667±0.2108*</td>
</tr>
<tr>
<td>MESA 200</td>
<td>3.837±0.4014**</td>
<td>1.500±0.2236*</td>
</tr>
<tr>
<td>MESA100</td>
<td>2.833±0.40141*</td>
<td>1.667±0.2108*</td>
</tr>
</tbody>
</table>

Values are mean ± SEM.  * P<0.05,  ** P<0.01,  Unmarked Not significant

One way ANOVA followed by Dunnett's test compared to control

Hole Board Test: The methanolic extract of Sarcostemma Acidum (Roxb.) Voigt, MESA200mg/kg and MESA 100mg/kg and standard drug Diazepam 2mg/kg, significantly increased the number of head pokes in hole board, when compared to control group (1% CMC). The standard drug showed maximum head pokes, then followed by MESA200 and the least pokes in MESA100, when compared to control.
Table. 2: Hole Board Test (Exploratory Behaviour).

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatment</th>
<th>Dose</th>
<th>No: of Head Dips</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Control CMC</td>
<td>1%</td>
<td>19.500±0.8466</td>
</tr>
<tr>
<td>II</td>
<td>Standard Diazepam</td>
<td>2mg/kg</td>
<td>32.167±1.138**</td>
</tr>
<tr>
<td>III</td>
<td>Test Methanol ext. SA(MESA)</td>
<td>200mg/kg</td>
<td>25.167±0.9098 **</td>
</tr>
<tr>
<td>IV</td>
<td>Test Methanol ext. SA(MESA)</td>
<td>100mg/kg</td>
<td>22.167±1.352</td>
</tr>
</tbody>
</table>

Values are mean±SEM. * P<0.05, ** P<0.01, Unmarked Not significant

One way ANOVA followed by Dunnets test compared to control.

Figure. 3.

Locomotor activity-Acto photometer. (CNS inhibitory activity)

The locomotor activity showed a decrease in standard and test groups. There was about 16% decrease in locomotor activity for the standard group and MESA 200 test compared to control. The test MESA200 and MESA 100 exhibited CNS inhibitory activity.

Table. 3. Locomotor Activity (Actophotometer).

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatment</th>
<th>Dose</th>
<th>Locomotor activity</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Control CMC</td>
<td>1%</td>
<td>148.50±3.810</td>
</tr>
<tr>
<td>II</td>
<td>Standard Diazepam</td>
<td>2mg/kg</td>
<td>124.50±2.540**</td>
</tr>
<tr>
<td>III</td>
<td>Test Methanol ext. SA(MESA)</td>
<td>200mg/kg</td>
<td>123.50±2.860**</td>
</tr>
<tr>
<td>IV</td>
<td>Test Methanol ext. SA(MESA)</td>
<td>100mg/kg</td>
<td>128.50±2.525**</td>
</tr>
</tbody>
</table>

Values are mean±SEM. * P<0.05, ** P<0.01, Unmarked Not significant

One way Anova followed by Dunnets test compared to control.
Catalepsy test-Anti Psychotic activity (Bar test).

The results of the bar test showed that the test groups MESA200 and MESA100, exhibited higher degree of catalepsy from 30minutes to 120minutes when compared to control. MESA200 possess maximum antipsychotic activity.

Table. 4. Catalepsy Test for Psychopharmacological Activity.

<table>
<thead>
<tr>
<th>Group</th>
<th>0 MIN n=6</th>
<th>30 MIN n=6</th>
<th>60 MIN n=6</th>
<th>90 MIN n=6</th>
<th>120 MIN n=6</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0.00</td>
<td>0.033±0.02108</td>
<td>0.01±0.016</td>
<td>0.033±0.02108</td>
<td>0.033±0.02108</td>
</tr>
<tr>
<td>Haloperidol 5 mg/kg</td>
<td>0.1167±0.1667**</td>
<td>4.784±0.2561**</td>
<td>4.9±0.1461**</td>
<td>5.133±0.0802**</td>
<td>4.467±0.1406**</td>
</tr>
<tr>
<td>MESA 200</td>
<td>0.0667±0.02108**</td>
<td>1.508±0.105**</td>
<td>1.717±0.07032**</td>
<td>1.867±0.1202**</td>
<td>1.933±0.06146**</td>
</tr>
<tr>
<td>MESA100</td>
<td>0.033±0.02108</td>
<td>1.333±0.08433**</td>
<td>1.400±0.06325**</td>
<td>1.3±0.0683**</td>
<td>1.283±0.0771**</td>
</tr>
</tbody>
</table>

Values are mean±SEM. * P<0.05, ** P<0.01, Unmarked Not significant

One way ANOVA followed by Dunnets test compared to control.
DISCUSSION
The whole dried plant of Sarcostemma acidum (Roxb) voigt was subjected to phytochemical analysis followed by pharmacological studies of methanolic extract.

In the present study the methanolic spray dried extract was used for Psychopharmacological studies. Psychopharmacological drugs are gaining importance in the present scenario and they have marked side effects. Herbal medicines for psychiatric disorders may possess fewer side effects.$^{[9,10]}$

Reduction in the locomotor activity of animals when tested using an Actophotometer indicates the C.N.S depressant property of the drug. The percentage change in the locomotor activity was about 16% lesser than the control group which indicates the C.N.S depressant property for MESA 200 and MESA 100.

Elevated plus maze test is used for screening drugs having anxiolytic effects$^{[11]}$ The number of entries into the open arm of EPM and time spent in the open arm were higher for MESA 200 and MESA 100 compared to control group, which indicates that the methanol extract possess anxiolytic like effect. This may be due to modulation of GABA receptors.

After administering the methanolic extracts to the rats, the number of nose poking into the holes in the Hole board apparatus were increased. The anxiolytic effect and exploratory behaviour is exhibited here. Catalepsy is a state of behavioural immobility. Antipsychotic drugs increases catalepsy. It is an indication of extrapyramidal side effects of antipsychotic drugs. Haloperidol induced catalepsy is an animal model to test of extrapyramidal side effects$^{[12]}$ this may be due to blockade of dopamine receptors in the brain. BAR test is done for catalepsy testing. The test revealed that the methanol extract increased the cataleptic activity.

CONCLUSION
The findings revealed the anti-anxiety activity, CNS depressant activity exploratory behaviour pattern changes and cataleptic activity of methanolic extract of the plant Sarcostemma Acidum (ROXB) voigt. It may be concluded that methanol extract of the whole plant possess the characteristics of the psychoactive group of drugs at the test dose levels. Further, the bioactive compound may be isolated in sufficient quantity so that it can be used for pharmacological studies.
ACKNOWLEDGEMENTS
The author is thankful to the Principal Department of Pharmacy M.G University Cheruvandoor for providing facilities for animal experiments.

REFERENCES