

Anxiolytic activity of Sangu Sunnam prepared both by Traditional and Modern Methods

R.SaravanaBhavan, R.Mallikeshwari, N.Chidhambranathan, Dr.B. jaiprakash

Dept. of Pharmacognosy, K.M College of Pharmacy, Utthagudi, Madurai-107

Dept. of Pharmacology, K.M College of Pharmacy, Utthagudi, Madurai-107

Dept. of Pharmacognosy, Kripnidhi College Bangalore – 34

In today's life of stress and strain there is a dire need for agents having neuro protective and neuropharmacological activity enhancing learning and memory function of the brain ^[1]. Stress involves complex biochemical, neural and immunological mechanisms and plays a crucial role in the genesis/progression of a variety of disease states ranging from psychiatric disorders, Cardio vascular disease, hypertension, peptic ulcers, migraine, allergies, asthma, carcinoma, pre mature ageing, rheumatic diseases and ulcerative colitis ^[2]. During the last two decades, pharmacotherapy with psychoactive drugs has been increasingly recognized as most effective in the management of anxiety, stress and psychosomatic disorders. However the prolong use of tranquilizers and psychotropic drugs leads to a variety of autonomic, endocrine, allergic, heamatopoietic and neurological side effects. The ancient system of medicine (Ayurveda and siddha) is a treasure of medicinal plants and calcined metals/minerals as remedies, to prevent and treat diseases. Hence in proposed study we have taken calcined Conch shell (Sangu Sunnam prepared both by traditional and modern methods) for its anxiolytic activity.

Key words: sunnam, neuropharmacology, anxiolytics

INTRODUCTION

In today's life of stress and strain there is a dire need for agents having neuro protective and neuropharmacological activity enhancing learning and memory function of the brain ^[1]. Stress involves complex biochemical, neural and immunological mechanisms and plays a crucial role in the genesis/progression of a variety of disease states ranging from psychiatric disorders, Cardio vascular disease, hypertension, peptic ulcers, migraine, allergies, asthma, carcinoma, pre mature ageing, rheumatic diseases and ulcerative colitis ^[2]. During the last two decades, pharmacotherapy with psychoactive drugs has been increasingly recognized as most effective in the management of anxiety, stress and psychosomatic disorders. However the prolong use of tranquilizers and psychotropic drugs leads to a variety of autonomic, endocrine, allergic, heamatopoietic and neurological side effects. The ancient system of medicine (Ayurveda and siddha) is a treasure of medicinal plants and calcined metals/minerals as remedies, to prevent and treat diseases. Hence in proposed study we have taken calcined Conch shell (Sangu Sunnam prepared both by traditional and modern methods) for its anxiolytic activity.

MATERIALS AND METHODS

ANIMALS: Wistar strain rats of either sex, weighing 180-225g were used for the study. Animals were housed in colony caged at an ambient temperature of $25 \pm 2^\circ\text{c}$ and 45-55% relative humidity with free access to food and water. All experiments were carried during light period (8.00-16.00h).

DRUGS AND CHEMICALS

Sangu sunnam and diazepam (calmpose 5mg/tab, Ranbaxy laboratories, India) were used as reference test and standard drugs in this study. Diazepam was administered in the form of suspensions using 1% CMC as the suspending agent. Sangu sunnam was administered with ghee preparation through orally. Diazepam is a renowned anxiolytic and anti-stress agent.

ASSESSMENT OF ANXIOLYTIC ACTIVITY IN RATS USING THE ELEVATED PLUS MAZE

Elevated maze is the simplest apparatus to study the anxiolytic response of all most all types of anti anxiety agents. The maze consists of 2 open arms (50cm x 10cm) crossed with 2 enclosed arms of the same dimensions with walls 40 cm height. The arms were connected with central square, 10cm x 10cm to give the apparatus a plus sign appearance. The maze was elevated 70cm above the floor in a dimly lit room ^[3]. Rodents have a natural aversion for high and open spaces and prefer enclosed arms, which have burrow like ambience and

TABLE NO. 1: EFFECT OF SANGU SUNNAM AND DIAZEPAM ON ANXIETY INDUCED IN RATS USING ELEVATED PLUS MAZE APPARATUS

| Treatment | Dose | Preference % Open arm | Time spent (s) Mean \pm SEM Open arm | No. of entries (Mean \pm SEM) Open arm |
|-------------|-----------------------------|--------------------------|----------------------------------------------|------------------------------------------------|
| Group – I | 10 ml / kg Normal Saline | 17.10 | 45.30 \pm 8.26 | 2.10 \pm 0.30 |
| Group – II | Diazepam 4mg/kg | 67.60 ^{*a} | 110.66 \pm 6.10 ^{*a} | 5.06 \pm 0.40 ^{*a} |
| Group – III | Sangu sunnam - T | 27.50 ^{*a} | 53.60 \pm 7.50 ^{*a} | 2.4 \pm 0.50 ^{*a} |
| Group - IV | Sangu sunnam - M | 53.30 ^{*a} | 75.10 \pm 7.50 ^{*a} | 3.10 \pm 0.65 ^{*a} |

T – Traditional Method M – Modern Method

- Values are expressed as Mean \pm SEM
- Values are find out by using ONE WAY ANOVA followed by Newmann Keul's Multiple range test
- ^{*a} values are significantly different from control at p<0.01

TABLE NO.2: EFFECT OF SANGU SUNNAM AND DIAZEPAM ON LOCO MOTOR ACTIVITY IN RATS USING ACTOPHOTOMETER APPARATUS

| Treatment | Dose | Loco motor activity (Scores) in 10 min (Mean \pm SEM) | | |
|-------------|-----------------------------|------------------------------------------------------------|--------------------|------------------------------|
| | | Before | After treatment | Reduction in activity (%) |
| Group – I | 10 ml / kg Normal Saline | 210 \pm 31.6 | 194.10 \pm 27.60 | ----- |
| Group – II | Diazepam 4mg/kg | 238.60 \pm 18.60 | 112.60 \pm 15.40 | 42.02 % ^{*a} |
| Group – III | Sangu sunnam - T | 198.50 \pm 17.60 | 174.55 \pm 12.45 | 10.00 % |
| Group - IV | Sangu sunnam - M | 195.50 \pm 20.50 | 156.50 \pm 10.50 | 19.00% |

T – Traditional Method M – Modern Method

- Values are expressed as Mean \pm SEM
- Values are find out by using ONE WAY ANOVA followed by Newmann Keul's Multiple range test
- ^{*a} values are significantly different from control at p<0.01

therefore spend greater amount of time in the enclosed arm. When exposed to the novel maze alley, the animals experience an approach-avoidance conflict, which is stronger in the open arms as compared to enclosed arms.

TREATMENT PROTOCOL:

The animals were divided into 4 groups; each group consists of 6 animals.

GROUP-I - Served as normal control received 10ml/kg Normal saline orally.

GROUP-II – Served as positive control, received 4mg/kg diazepam, suspended with 1ml of 1% CMC, orally.

GROUP-III- Served as treatment control, received 3mg/kg of sangu sunnam(traditional method)dissolved with 4ml of ghee orally.

GROUP- IV- Served as treatment control, received 3mg/kg of sangu sunnam(Modern method)dissolved with 4ml of ghee orally.

One hour following drug administration each rat was placed individually in the corner of an open arm and observed for a period of 10 min^[4]. The parameters noted was

i) Percent performance for open arms as first entry.

ii) Number of entries in the open arm

iii) Duration of stay in the open arm

Increased exploratory activity in the open arm was an indication of anxiolytic activity.

ASSESSMENT OF ANTI-DEPRESSANT ACTIVITY IN RATS USING AN ACTOPHOTOMETER.

LOCOMOTOR ACTIVITY

The loco motor activity (horizontal activity) was measured using an actophotometer. The movement of the animal cuts off a beam of light falling on a photo cell and a count was recorded and displayed digitally. Each rat was placed individually in the actophotometer for 10 min and basal activity scores were recorded. Then the animals were divided into 5 groups each having 6 animals.

TREATMENT PROTOCOL:

GROUP-I - Served as normal control received 10ml/kg Normal saline orally.

GROUP-II – Served as positive control, received 4mg/diazepam, suspended with 1ml of 1% CMC, orally.

GROUP-III-Served as treatment control, received 3mg/kg of Sangu Sunnam (Traditional method) dissolved with 4ml of ghee orally.

GROUP- IV- Served as treatment control, received 3mg/kg of Sangu Sunnam (Modern method) dissolved with 4ml of ghee orally.

One hour following drug administration each rat was placed again in the actophotometer for recording the activity scores as described earlier^[6].

RESULTS

Evaluation of anxiolytic activity using elevated plus maze apparatus

The elevated plus maze comprising of two open and two enclosed arms, produced a novel environment which helped in inducing anxiety in animals, because of the open nature of the arms and elevation (70cm) from the floor. When the animals were placed on the maze, they showed a preference for the enclosed (dark) arms and showed anxiety and fear like movements characterized by immobility, freezing and defecation on entering the open arms. The Sangu sunnam by both methods at doses (3mg/kg) produced significant increase in percent preference for open arm as first entry, in total number of entries in the open arm, as well as in the duration of stay in the open arm, when compared to the control group.(Table No-1). Diazepam (4mg/Kgp.o) significantly increased ($P < 0.001$) the percent preference for open arm, the number of entries as well as the duration of stay in the open arm, indicating anxiolytic activity.

Assessment of Locomotor Activity

The Sangu sunnam by both methods at doses (3mg/kg) produce no significant reduction in locomotor activity as compared to the control animals receiving normal saline. (Table No. 2) However the diazepam treated group revealed a statistically significant decrease in locomotor activity as compared to the control.

DISCUSSION

In our investigation, the Sangu sunnam by both methods at doses (3mg/kg) produces significant change in the exploratory activity of the rats in the elevated plus maze model. Anxiolytic compounds, by decreasing anxiety, increase the open arm exploration time as well as the number of entries into the open arm.

Generally most of the anxiolytic agents have an adverse effect on memory as seen with the benzodiazepines, commonly used as anxiolytics⁷. An important point to be noted is that recently the plus maze model is also being used to study learning and memory induced by scopolamine, an anti-cholinergic agent, is reflected by prolonged transfer latency from the open arm to the closed arm⁸. With respect to our findings, The Sangu sunnam by both methods at doses (3mg/kg) causes an increase in the number of entries into the open arm. It could thus also be inferred that the rats retain the memory of the aversive quality of the open arm and this could probably be considered a significant finding with respect to the sangu preparation. Locomotor activity is considered as an index of alertness and a decrease in it would indicate sedative activity. However, the Sangu sunnam by both methods at doses

(3mg/kg) was found to have no effect on the Locomotor activity. Moreover, the lack of effect on Locomotor activity works to the advantage of the sangu sunnam demonstrating nootropic activity.

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