Neuroprotective potential of *Costus speciosus* on neurotransmitters levels in noise stress condition in rats

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**ABSTRACT**

Exposure to overwhelming stress could lead to development of mental disorders and worsening of existing ones. The impact produced by exposure to stress might be affected by the seriousness of the stressor, the time amid which it is connected (acute vs. chronic). In traditional practice of medicine, *C. speciosus* extract exhibits antistress action so we aimed to test whether this extract plays a vital role in maintaining the brain’s chemical transmitters levels during acute noise stress conditions. The levels of Dopamine (DA), Norepinephrine (NE) and Serotonin (5HT) were analysed by reverse phase HPLC in cerebral cortex and hippocampus brain tissue of acute noise stress exposed, *C. speciosus* extract (250mg and 500mg) and fluoxetine treated rats. Norepinephrine and dopamine were found to be increased in contrast to 5-HT levels in stressed group of rats, whereas CSE 500 mg could alleviate the said alteration. This study highlights the importance of *C. speciosus* extract therapy to noise stress induced rats by testing the neurotransmitters levels in brain tissue to understand its therapeutic potential.

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**INTRODUCTION**

Stress is experienced by everybody and can affect the physiological functions of an individual (Britz and Pappas, 2010). Noise stress can cause an elevation of blood pressure, headaches, trouble sleeping, inhibit the immune system and have an overall negative impact on an individual’s physical and mental well-being (Britz and Pappas, 2010). Elevated amounts of noise can be related with debilitation of capacity to focus. It has appeared to create various physiological, biochemical, and neurochemical rejoinder in both human and animals. Intense and perpetual presentation to boisterous noise instigates oxidative stress that produces extreme free radicals (Bajwa et al., 2014). The noise stress causes the neurosignals interruption due to alteration in neurotransmitters levels. It was observed that many herbal medicines may have a neuroprotective effect to prevent the neurosignal interruptions.

*C. speciosus* is among the most effective Islamic traditional medicinal plants. Traditionally *C. speciosus* was used as a remedy for pharyngitis, tonsillitis, pleurisy and antidote for snake venom. *C. speciosus* rhizomes and roots are prove to have anthelmintic, anti-inflammatory, antidiabetic, hepatoprotective,
antihyperlipidemic, antispasmodic and antimicrobial activities (Saraf, 2010). Moreover, C. speciosus leaf is used for patients with high fever. Rhizome juice is given to treat leprosy and for headache relief (El-Far, 2018). Thus, the anti-inflammatory activities exhibited by the isolated compounds from C. speciosus serve as a promising and expanding strategy for treatment of various inflammatory disorders and related symptoms.

Based on the observed review this plant is effective in the management of stress diseases, so we evaluated the effect of ethanol extract of the rhizomes of C. speciosus on neurotransmitters levels in acute noise stress exposed rats.

MATERIALS AND METHODS

Plant collection

The plant was obtained from a local nursery in Chennai. After authentication by Plant Authentication and Research Centre, Tambaram, Chennai, Tamil Nadu and the plant was cultivated.

Plant extract preparation

The rhizomes of Costus speciosus were cleaned, shade dried and powdered using an electrical grinder and was passed through sieve no. 40 for removing the debris. The sieved powder was kept (37ºC) in an airtight container for further use. Ethanol extract of the powder was prepared by using soxhlet apparatus. The extracts were filtered and the filtrate was heated at 70ºC so that alcohol gets evaporated and a semisolid product was obtained. This semisolid product was converted to powder form by a flash evaporator. The final product was stored until further use. The extract was administered to the animals orally using a gavage after mixing with 5% Tween 80 as a vehicle.

Noise stress induction procedure

The animals were kept in a noise stress chamber that was fabricated exclusively for this kind of study. A pure tone noise of sine waves was created with the help of a function generator. Using an amplifier, these sine waves were amplified. The amplifier was attached to a full-range loud speaker and was kept at the roof of the chamber at a height of 40 cm above animal cage. For obtaining uniform noise in the whole area of the chamber, a column speaker was used. A pure tone noise of sine waves with a frequency of 10 kHz and an intensity of 100 dB were used. A decibel meter was used for measuring and maintaining the noise intensity (Sembulingam et al., 2000). In acute study, the animals were subjected to 30 min of noise stress. Before the beginning of the experimental procedure, all the animals were subjected to an auditory test by using finger clicking method.

Experimental design

Male wistar strain albino rats (n=60) weighing 150-180 g was taken for this study. The animals were classified into five groups, each having six animals to study the acute noise stress effects.

Acute noise stress study

Group I: Control group used for determining the baseline values of each parameter of the study; Group II: Acute stress group subjected to acute noise stress for 30 mins. Group III: Standard drug treated group. The animals pre-treated with the 10mg/kg body weight of fluoxetine for 7 days. On the 8th day, these rats were exposed to noise stress for 30 minutes. Group IV: The animals pre-treated with the Costus speciosus extract (CSE) 250mg/kg body weight orally for 7 days and then exposed to noise stress for 30 minutes; Group V: The animals of this group pre-treated with the CSE (500mg/kg body weight) orally for 7 days and then exposed to noise stress for 30 minutes.

Preparation of brain tissue homogenate

Brain samples were then homogenized in extraction medium. Extraction medium was made by perchloric acid (0.4 M) with sodium metabisulphite (0.1%), EDTA (0.1%) and cysteine (0.01%). Samples were then centrifuged at 10000 × g in eppendorf tubes for 15 minutes at 4°C. Supernatants were used for HPLC analysis using fluorescent detector.

Estimation of neurotransmitters

The levels of Dopamine (DA), Norepinephrine (NE) and Serotonin (5HT) were isolated by reverse phase HPLC with fluorescent detection. The isolation was carried out on the reversed-phase C18 column by a gradient elution using acetonitrile as an eluent. The column was equilibrated with the initial mobile phase for 10 min before injecting the next sample. The flow rate was constant at 1.0 mL/min and the column temperature was fixed at 30°C. The fluorescence excitation were set at λ maximum 333 nm and emission wavelengths were set at λ maximum 390 nm, respectively. The derivatives separated were quantified by fluorescence detector.

RESULTS AND DISCUSSION

Neuroprotection by Costus speciosus has been reported in several in vitro studies and in vivo studies (Swati et al., 2015). It has been shown to protect neurons from oxidative damage during noise stress condition and its phytoconstituents efficacy acts
as a lead compound in clinical applications. Based upon the previous findings we tested this plant efficacy on neurotransmitters levels in cortex and hippocampus region, during acute noise stress environment.

Figure 1: Effect of CSE treatment on 5-hydroxytryptamine levels in cortex and hippocampus tissue of acute stress induced rats.

Figure 2: Effect of CSE treatment on dopamine levels in acute noise stress induced rats.

Figure 3: Effect of CSE treatment on noradrenaline (NE) levels in acute noise stress induced rats.

The 5-hydroxytryptamine (5-HT) and dopamine (DA) chemicals have a major role in learning and memory functions. The increased 5-HT and dopamine levels could be the result of the deleterious activities of noise on the hippocampus (Jayakumar, 2017). In the current study 5-hydroxytryptamine was found to be increased in hippocampal and cortex tissue of rats exposed to noise stress. A significant (p<0.05) decrease in 5-HT levels was observed in CSE (500mg) and fluoxetine treated rats (Figure 1) after the exposure of noise, whereas non-significant changes were observed in 5-HT levels both in cortex and hippocampus region upon treatment with CSE (250mg). Results were expressed as Mean ± SEM (n=6). *p<0.05 statistically significant as compared with normal control group. #p<0.05, “p<0.05 statistically significant as compared with noise stress group. NS- Noise stress; NS+ Flux- Noise Stress+ fluoxetine 10mg/ kg; NS+ CSE 250- Noise Stress + Costus speciosus extract 250mg / kg; NS+CSE 500 - Noise Stress + Costus speciosus extract 500mg/kg.

The stress-induced stimulus of serotonergic and dopaminergic systems has been observed as plasticity that aids the brain reorganize its neuronal network (Hasegawa, 2018). The increase in DA levels in cortex and hippocampus tissue of noise-exposed rats (Figure 2) may reduce the plasticity of synapses resulting in impairment of memory. The present study suggests that changes in neurotransmitter levels following exposure to acute noise stress would be useful to better understand the mechanism involved in noise-induced behavioral deficits and help to know the C. speciosus therapeutic strategies to combat the harmful effects of noise. The dopamine levels was decreased in CSE (250 mg) and (500 mg) treated rats in contrast to fluoxetine treated rats in cortex region and in hippocampus region non-significant changes noted in neurotransmitters levels as compared with stress exposed rats. Results were expressed as Mean ± SEM (n=6). *p<0.05 statistically significant as compared with normal control group. #p<0.05 statistically significant as compared with noise stress group.

A variety of stresses produces a significant alteration in various neurotransmitters in the central nervous system (CNS) as well as peripheral nervous system, which causes depletion of norepinephrine level in the brain (Haenisch et al., 2009). It appears that norepinephrine is utilized in response to stress, which leads to increase in dopamine concentrations. Norepinephrine was found increased in stressed group of rats significantly (p<0.001) compared to normal control, whereas CSE 500 mg could alleviate the alteration in NE among stressed rats (Figure 3). The minimal variation was noted in fluoxetine treated groups and no significant changes noted in CSE(250mg) treated rats in the cortical
tissues and in hippocampus region also after the exposure of noise. Results were expressed as Mean ± SEM (n=6). ***p<0.05 statistically significant as compared with normal control group. *p<0.05, "p<0.05 statistically significant as compared with noise stress group. C. speciosus extract significantly reduced the stress-induced rise of 5-HT, dopamine and norepinephrine levels in brain tissues by preventing the alarm reaction which may be due to its antidepressant effect (Verma and Khosa, 2017).

CONCLUSIONS

The ethanol extract of rhizomes of Costus speciosus could alleviate acute noise stress induce alterations in neurotransmitters release in cerebral cortex and hippocampus. Since C. speciosus has a good effect comparative to the standard drug fluoxetine it can serve as an add-on drug to current regimens.

Conflict of Interest

The authors declare that they have no conflict of interest for this study.

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