Anti-stress Activity of *Mitragyna africanaus* (Willd), Stembark Extract

B. M. Aji, K. D. Effraim and I.P. A. Onyeili

The acute toxicity and the effect of *Mitragyna africanaus* stembark extract of methanol (CH₃OH) on stress were investigated in rats, using its effect on pentylenetetrazole induced convulsions and its muscle relaxant effect. The extract did not produce any death in the treated rats even at the highest dose (6400mg kg⁻¹) used. It did not protect rats treated with convulsive doses of pentylenetetrazol (100mg kg⁻¹) but increased the period of onset of convulsions and decreased the number of spasms. It also showed significant (p<0.05) muscle relaxant effect.

Keywords: *Mitragyna africanaus*, acute toxicity, pentylenetetrazol, muscle relaxant, anti stress activity

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Introduction

*M. africanus* belonging to Rubiaceae family is a large tree widely distributed in Nigeria. It is known among the natives as "uburu" in Igbo; "abara" in Yoruba; "gysaya" in Hausa. The bark and the leaves are used in West Africa for the treatment of mental disorder and epilepsy (von Maydell, 1990). The extract from this plant is usually used as a decoction either alone or with other plant materials. The decoction of the stem bark mixed with Garcinia cola seed extract is used in South Eastern Nigeria for African sleeping sickness (trypanosomiasis). The plant is listed by (Walker, 1953) as one of the ingredients in the treatment of sterility, when it is combined with other plant materials such as *Coulia edulis*, *Isolana lecti*, *Bartiera fistulosa* and *Alchornea cardifolia* in a decoction by the Bupunus of Gabon. In the North Eastern Nigeria, it is combined with *Ficus thonningii* (stem bark) and *Ziziphus spinosa-christi* (stem bark) for treatment of mental illness (Abdulrahman, 1982). In earlier studies Mitragyna, alkaloids have been observed to have a local anesthetic properties (Oliver-Sayer, 1986; Annoa, 1986; Ajii, 1998). The objective of this work is to investigate the effects of stem bark extract of *M. africanus* on stress by using experimental rats.

Materials and Methods

Plant Identification and Collection: The *M. africanus* was identified by the Department of Biological Sciences, University of Maiduguri. The stem bark was collected in the month of May 1999, and was air dried at a voucher herbarium (Ajii) for reference was deposited in the Department of Biological Science, University of Maiduguri.

Preparation of the Extract: The dried stem bark was powdered and 50g of it was extracted by boiling with 350ml of 80% methanol (CH3OH) for 15 min and allowed to cool. It was then filtered and the filtrate was concentrated in a water bath at a temperature of 80°C for 7 hours. The concentrated extract was stored at 4°C until used.

Test Animals: Wistar rats (150 - 160g) of both sexes were purchased from National Veterinary Research Institute, Vom (Jos, Nigeria) and were housed in cages. They were given food (ECWA Feed, Nigeria Ltd, Maiduguri) and water ad libitum, they were allowed to adjust to the laboratory environment for one week before the commencement of experiment.

Acute Toxicity Testing: Thirty-five wistar rats were used and they were randomly separated into seven group (5 rats each) and were allowed to free access of food and water. The animals in groups 1 to 7 were injected intraperitoneally with various doses (100, 200, 400, 800, 1600, 3200 and 6400mg kg⁻¹) of *M. africanus* extract in distilled water respectively. The symptoms of toxicity in each rat were observed, and the number of rats that died within 24 hr was also recorded.

**Anti-stress Effect using Pentylentetrazol induced Convulsions:** A convulsive dose (100mg kg⁻¹) of Pentylentetrazol was given subcutaneously to the 5 rats of each of two groups (A and B). Rats in group B were however, pre-treated with a therapeutic dose (400mg kg⁻¹) i.p.i. of the stem bark extract 30 min, before treatment with the convulsant. The onset of convulsions, number of convulsions per min and duration of convulsions were recorded (Takagi et al., 1963; Maeda, 1981). The results obtained were analyzed by one-way analysis of variance, (ANOVA).

Muscle Relaxant activity by Inclined Board Method: The method of Kitano et al. (1985) was used. Twenty rats of both sexes were divided into four group (A, B, C, D). The rats were placed one after another on the smooth surface of a board inclined at 36° to the horizontal before and 30 min after treatment with varying doses of the extract (60, 100, 200 and 400mg kg⁻¹ respectively) intra-peritoneally and allowed a minimum of 10 seconds to remain on the board. The rats that slipped down the board before 10 seconds were counted as positive for muscle relaxation.

Results

The methanol extract of *M. africanus* stem bark was sticky and dark brown in colour. The yield of the extract was 20% (w/v).

Acute Toxicity: The clinical symptoms observed in rats following administration of methanol stem bark extract of *M. africanus* include depression, hind limb paralysis, recumbency, sleeping and difficulty in respiration, which was enhanced with increasing dose of the extract. The effect of the extract lasted for 8-12 hours, following which, the rats returned to normal activity. No mortality was recorded in any of the treated groups hence the LD₅₀ could not be calculated (Table 1).

**Anti-stress Effect on Pentylentetrazol induced convulsion:** The stem bark extract of *M. africanus* did not protect rats treated with convulsive dose of pentylentetrazol, however, the mean number of spasms per min. was reduced by 54%, and the mean onset of convulsion was increased by 46%. The mean time lapse between convulsion and death was increased by 22% (Table 1). All rats treated with pentylentetrazol i.e. (group A = control, B = pretreated with *M. africanus* died.

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<th>Table 1: The Effect of <em>M. africanus</em> stem bark extract in rats treated with convulsive dose (100mg /kg S.C) of pentylentetrazol</th>
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<th>Table 2: Effect of <em>M. africanus</em> stem bark extract on muscle relaxation (inclined board method) in rats</th>
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Muscle Relaxant Activity: None of the rats treated with 50mg kg⁻¹ of the extract slid down the board, while 40 and 80% of rats treated with 100 and 200mg kg⁻¹ of the extract respectively slid down the board. There was 100% muscle relaxation with 400mg kg⁻¹ of the extract (Table 2). The muscle relaxant activity of the extract appears to be dose dependent.

Discussion
The methanol extract of the stembark of M. africanus showed profound anti-stress activity, since the extract appeared to be a sedative action; some rats dosed with the extract even went to sleep. The extract did not protect rats treated with a convulsive dose of pentylenetetrazol, however, it reduced the mean number of spasms per min. by 54%, and increased the mean onset of convulsion by 46%. The inability of the extract to prevent the convulsion may be due to the low dose used for the protection of the animals against pentylenetetrazol. Most anti-convulsant activity of plant extracts were based on the ability of those extracts to delay the onset of seizures (Yidya et al., 1990; Diwan et al., 1991; Martins and Rao, 1991). This extract also possess muscle relaxant activity, as shown by its effects in the inclined board test which can evaluate the muscle relaxant activity (Kasahara and Hikino, 1987).

In conclusion, the stembark extract of M. africanus showed anti-stress effect by inducing depressant effect on the nervous system. It showed muscle relaxant activity and reduced the mean number of spasms per min. as well as increase the mean onset of convulsion but did not protect rats from convulsion induced with pentylenetetrazol.

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References


