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Original Article

Cymbopogon citratus aqueous leaf extract attenuates neurobehavioral and biochemical changes induced by social defeat stress in mice

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ABSTRACT

Objective: Psychosocial stress has been implicated in the genesis of psychiatric disorders such as memory deficits, depression, anxiety and addiction. Aqueous leaf extract of *Cymbopogon citratus* (CYC) otherwise known as lemongrass tea has antidepressant, anxiolytic and anti-amnesic effects in rodents. This study was designed to evaluate if *C. citratus* could reverse the neurobehavioral and biochemical derangements induced by social defeat stress (SDS) in the resident/intruder paradigm.
Methods: Intruder male mice were divided into five groups (n = 7): group 1 received saline (10 mL/kg, p.o.; non-stress control), group 2 also received saline (10 mL/kg, p.o.; SDS control) while groups 3–5 had *C. citratus* (50, 100 and 200 mg/kg, p.o.) daily for 14 d. The SDS was carried out 30 min after each treatment from day 7 to day 14 by exposing each intruder mouse in groups 2–5 to a 10 min confrontation in the home cage of an aggressive resident counterpart. The neurobehavioral features (spontaneous motor activity-SMA, anxiety, memory, social avoidance and depression were then evaluated. The concentrations of nitrite, malondialdehyde and glutathione as well as acetylcholinesterase activity in the brain tissues were also determined.
Results: *C. citratus* (50, 100 and 200 mg/kg) attenuated hypolocomotion, heightened anxiety, depressive-like symptom, memory deficit and social avoidance induced by SDS. The altered levels of oxidative stress and acetyl-cholinesterase in SDS-mice were positively modulated by *C. citratus.*
Conclusion: The results of this study suggest that *C. citratus* might mitigate psychosocial stress-induced neurologic diseases in susceptible individuals.
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1. Introduction

Psychosocial stress has been highlighted as the major source of stressful life event encountered by human regularly on daily basis (Kanarik et al., 2001). Most importantly, there is countless number of potential stressors of psychosocial domain in our world today; existing in workplaces, schools, families/home settings and the society at large (Björkqvist, 2001; Solomon, 2017). Joblessness, marital problems, poverty or economic pressure, violence, insecurity, caring for an ailing parent or disabled child among many other factors have been identified as chronic psychosocial stressors (Björkqvist, 2001; Solomon, 2017). Besides, if the stress persists and becomes chronic, low self-esteem, social maladjustment and various psychopathologies may ensue (Björkqvist, 2001; Kessler, 2003). Thus, psychosocial stress has been implicated for the development of several psychiatric pathologies such as social withdrawal, memory decline, depression, anxiety and addiction (Gaurav, Solanki, Atrooz, Allam, & Salim, 2013). Furthermore, models based on social conflict that typify a social form of stress (social defeat stress) have also been reported to have clear advantages over environmental models for the elucidation of stress-induced psychopathology in humans (Krishnan et al., 2007). The resident-intruder paradigm has been described as a suitable model to study the effects of chronic social stress-induced phenotypes and endophenotypes relevant to psychiatric diseases (Solomon, 2017; Gaurav et al., 2013). The resident-intruder paradigm is based on intra-species territorial aggression and continuous subordination in males (Malick, 2010). It involves exposure of an intruder animal in the home-cage of an aggressive resident counterpart resulting in an attack and defeat of the intruder (Lio et al., 2012). Exposure to a single attack or defeat is known as acute social defeat stress whereas repeated exposures for consecutive days or weeks is referred to as chronic social defeat stress (Lio et al., 2012).

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Moreover, chronic social defeat stress has been shown to cause more damage to the brain through glucocorticoid-mediated induction and sustained oxidative stress and neuroinflammation as a result of deregulation of hypothalamic–pituitary adrenal (HPA) pathway (Krishnan et al., 2007; Gaurav et al., 2013).

Cymbopogon citratus (DC.) Stapf (lemongrass) is a valuable medicinal and nutritional plant found in most tropical countries. *C. citratus* is well recognized in many countries as flavoring agent and for treatment of infections, stomach aches and rheumatic pain in traditional medical practice (Carlini et al., 1986; Figueirinha, Paranhos, Perez-Alonso, Santos-buelga, & Batista, 2008). However, the aqueous leaf extract of *C. citratus* widely known as lemongrass tea is readily consumed for its alleged beneficial effects in persons with central nervous system ailments (Carlini et al., 1986). Previous pharmacological studies have established that lemongrass has sedative, anticonvulsant, antidepressant, anxiolytic and memory promoting effects (Carlini et al., 1986; Yang, Xi, Li, & Qu, 2009; Umukoro, Ogboh, Omorogbe, Adekeye, & Olatunde, 2017). The presence of essential oils (such as citral, borneol, estragole, methyleugenol, geranyl acetate, geraniol, beta-myrcene, limonenepiperitone, citronellal, citrat-2, alpha-terpineole, pinene, farnesol, proximadiol, and (+)-cymbodiacetal) and other bioactive compounds such as luteolin, isoscoparin, quercetin, kaempferol, and apigenin have been reported to contribute to the pharmacological activities demonstrated by *C. citratus* (Figueirinha et al., 2008; Yang et al., 2009). This present study was designed to evaluate the effects of the aqueous extract of the leaf of *C. citratus* on some neurobehavioral and biochemical derangements induced by SDS in mice.

2. Materials and methods

2.1. Laboratory animals

Male Swiss mice (25–27 g) used in this study were purchased from the Central Animal House, University of Ibadan, Ibadan and housed in plastic cages at room temperature. The animals had free access to rodent pellet diet and water ad libitum. They were acclimatized for two weeks to the laboratory conditions before commencement of the study. The animals were handled in accordance with the National Institutes of Health (NIH) Guidelines for the Care and Use of Laboratory Animals.

2.2. Collection of plant material and extraction procedure

The fresh leaves of *C. citratus* were collected from the Botanical Garden of the University of Ibadan, Ibadan, Nigeria. The leaves were washed, sun-dried and ground using electric blender. The aqueous leaf extract of *C. citratus* was prepared according to the procedure earlier described by Umukoro et al. (2017). Six hundred grams (600 g) of the powdered dried leaf material was soaked in distilled water for 24 h. The solution was then filtered using Whatman3 mm thick filter paper. The filtrate was concentrated using a rotary evaporator at 40°C and the residue was dried in a desiccator before being kept in a sterilized glass. The aqueous extract of *C. citratus* was made by dissolving it in normal saline immediately before use. The doses of 50, 100, and 200 mg/kg of *C. citratus* used in the study were chosen based on information obtained from previous investigations (Umukoro et al., 2017).

2.3. Drugs and chemicals

Acetylthiocholine, Ellman Reagent [5′5′-dithiobis-(2-nitrobenzo ate) DTNB] and thiobarbituric acid (TBA) were purchased from Sigma-Aldrich, St. Louis, USA. Trichloroacetic acid (TCA) was obtained from Burgoyne Burbidges & Co., Mumbai, India.

2.4. Procedure for induction of chronic social defeat stress

The social defeat stress (SDS) was carried out using the resident intruder model according to the procedure earlier described (Krishnan et al., 2007). Male resident mice were made aggressive by housing them individually with female counterparts for 3 weeks. However, the male mice that served as intruders were housed in groups and randomly divided into five experimental groups (n = 7). Mice in group 1, serving as non-stress control received saline (10 mL/kg, p.o.), group 2, also received saline (10 mL/kg, p.o.) but served as SDS control; groups 3–5 were treated with *C. citratus* extract (50, 100 and 200 mg/kg, p.o.) daily for 14 consecutive days. However, SDS was carried out 30 min after treatments from day 7 to day 14 by subjecting each intruder mouse in groups 2–5 to a 10 min confrontation in the home cage of an aggressive resident mouse. The intruders were subjected to social defeat stress from different aggressive residents each day, so as to enhance aggression of the residents and reduce familiarity between the resident and intruder on daily basis (Golden, Covington, Berton, & Russo, 2011). Social defeat was observed each day in the intruders as shown by upright defensive postures, submissive postures, flight and vocalizations. Mice were returned to their home cages after each social defeat session.

2.5. Effect of C. citratus on SDS-induced behavioral deficits

The effect of aqueous leaf extract of *C. citratus* on SDS-induced behavioral deficits such as altered spontaneous motor activity, memory deficit, anxiety, social interaction and depressive-like behaviors were evaluated in this sequence after the last session of SDS on day 14.

2.5.1. Test for spontaneous motor activity

The effect of aqueous leaf extract of *C. citratus* on SDS-induced changes in SMA was evaluated using activity cage. Mice in each group were placed individually in the activity cage and the SMA was recorded as activity counts per 5 min. The floor of the cage was cleaned with 70% ethanol after each test to prevent olfactory cue from previous animals.

2.5.2. Effect on memory performance

The Y-maze test was used to evaluate the effect of aqueous leaf extract of *C. citratus* on SDS induced memory impairment according to the procedure previously described (Casadesus et al., 2006). The Y-maze apparatus consists of three identical arms (A, B and C), which are symmetrically separated at 120°. Immediately after the open field test, the animals were placed individually in the Y-maze apparatus at the end of arm A and allowed to explore all the three arms freely for 5 min. The number of arm visits and sequence (alternation) of arm visits were then recorded. The percentage alternation, which is used as an index for spatial memory was, calculated (Casadesus et al., 2006). After each test, the apparatus was also cleaned with 70% ethanol to remove residual odor. Alternation behaviors were defined as consecutive entries into all three arms (i.e. ABC, CAB or BCA but not BAB) (Casadesus et al., 2006).

2.5.3. Light/dark box test

The light–dark box (LDB) was used to evaluate the effect of aqueous leaf extract of *C. citratus* on SDS-induced anxiety behavior according to the procedure earlier described (Bourin & Hascoet, 2003). The animals were placed individually at the centre of the bright compartment of the light/dark box apparatus and allowed to explore the apparatus for a period of 5 min. The total time spent (s) in the light and dark compartments were then recorded. The LDB was also cleaned with a solution of 70% ethyl alcohol in order to prevent odor bias.

2.5.4. Tail suspension test

The tail suspension test (TST) is commonly used as a screening procedure for evaluation of novel compounds with anti-depressant property in rodents (Steru, Chermat, Thierry, & Simon, 1985). After the light/dark box test, each mouse was then suspended 50 cm above the floor, with the help of an adhesive tape placed approximately 1 cm from the tip of the tail. The period of immobility (s) was measured for 4 min after the initial 2 min delay and the animals were considered immobile whenever they remained motionless and hung passively (Steru et al., 1985).

2.6. Brain tissue preparation for biochemical studies

Immediately after behavioral testing, the animals were euthanized under ether anaesthesia and the brains were removed, weighed and homogenized with 5 mL of 10% phosphate buffer (0.1 mol/L, PH 7.4). The brain tissue homogenate was centrifuged at 100 00 rpm at 4°C for 10 min and the supernatants were immediately frozen and stored until the different biochemical assays were carried out.

2.6.1. Glutathione (GSH) content

The procedure described by Jollow, Michell, Zampaglione, and Gillete (1974) was used to estimate the brain GSH content. Briefly, the brain homogenate (0.4 mL) was added to 0.4 mL of 20% trichloroacetic acid (TCA) and mixed by a gentle swirling motion. The resulting mixture was then centrifuged at 4°C for 20 min at 5400*g*. Then, 0.25 mL of the supernatant was added to 2 mL of 0.6 mmol/L 5,5′-dithio-bis(2-nitrobenzoic acid) (DTNB) and the final volume of the solution was made up to 3 mL with phosphate buffer (0.2 mol/L, pH 8.0). Absorbance was read at 412 nm against blank reagent (2 mL of 0.6 mmol/LDTNB + 1 mL phosphate buffer (0.2 mol/L, pH 8.0) using a spectrophotometer. The concentration of reduced GSH in the brain tissues were expressed as nanomoles per milligram protein (nmol/mg protein).

2.6.2. Malondialdehyde (MDA) levels

The brain content of MDA was measured according to the method previously described (Okhawa, Ohishi, & Yagi, 1979). Briefly, an aliquot of 0.4 mL of the brain tissue was mixed with 1.6 mL of Tris-potassium chloride (Tris-KCl) buffer to which 0.5 mL of 30% TCA was added. Then, 0.5 mL of 0.75% TBA was added and placed in a water bath for 45 min at 80°C. This was then cooled in ice and centrifuged at 1200*g* for 15 min. The clear supernatant was collected and absorbance measured against a reference blank of distilled water at 532 nm. The MDA level was calculated using the Molar extinction coefficient of 1.56 × 105 mol/L/cm and the value was expressed as nanomole of MDA per mg protein (nmol MDA mg^{-1} protein).

2.6.3. Acetyl-cholinesterase (AChE) activity

The procedure described by Ellman, Courtney, Andre, and Featherstone (1961) was used to estimate AChE activity. Briefly, the supernatant of the brain tissue (0.4 mL) was added to 2.6 mL of phosphate buffer (0.1 mol/L, pH 7.4) and followed by addition of 0.1 mL of DTNB. Then, 0.1 mL of acetylthiocholine iodide solution was added to the reaction mixture. The absorbance was read using a spectrophotometer at a wavelength of 412 nm and change in absorbance for 10 min at 2 min interval was recorded. The rate of acetyl-cholinesterase activity was measured following increased coloration produced from thiocholine when it reacts with DTNB. The change in absorbance per minute was determined and the rate of AChE activity was estimated and expressed as μmol/min/mg protein.

2.6.4. Protein content estimation

This assay was carried out according to the procedure described by Gornall, Bardawill, and David (1949) using the Biuret method. The diluted sample (1 mL) of the brain homogenate was taken and added to 3 mL of Biuret reagent in triplicate. The mixture was incubated at room temperature for 30 min after which the absorbance was read at 540 nm using distilled water as blank. Bovine serum albumin (1 mg/mL) was used as standard and was measured in the range of 0.01–0.1 mg/mL.

2.7. Statistical analysis

The data obtained were expressed as mean ± S.E.M (standard error of mean) and analyzed with Graph Pad Prism software version 5.00. Statistical analysis of data was done using One-way ANOVA, followed by Bonferroni post-hoc test. *P*-values less than 0.05 ($P < 0.05$) were considered statistically significant.

3. Results

3.1. C. citratus attenuates impaired locomotor activity induced by social defeat stress

The effect of *C. citratus* on SDS-induced impairment in locomotor activity was shown in Fig. 1. Mice subjected to SDS exhibited a significant ($P < 0.05$) decrease in locomotor activity when compared with non-stress control (Fig. 1). However, oral administration of *C. citratus* (50 and 200 mg/kg) significantly ($F [4,30] = 11.22$, $P < 0.05$) attenuated decreased locomotor activity induced by SDS.

3.2. C. citratus reduces anxiety-like behavior in mice exposed to social defeat stress

The effect of *C. citratus* on SDS-induced anxiety-like behavior was shown in Fig. 2. Social defeat stress produced a significant ($P < 0.05$) increase in anxiety-like behavior as evidenced by increased time spent in the dark compartment when compared with non-stress group. But, *C. citratus* extract (100 and 200 mg/kg, p.o.) significantly ($P < 0.05$) reduced the anxiety-like behavior in mice subjected to SDS.

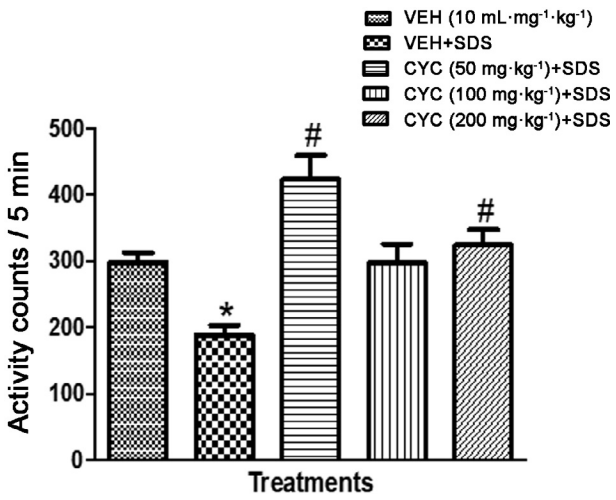

Fig. 1. Effect of aqueous leaf extract of *C. citratus* (CYC) on social defeat stress (SDS)-induced impaired locomotor activity in mice (mean ± S.E.M, $n = 7$). $^{*}P < 0.05$ *vs* vehicle (VEH) group; $^{\#}P < 0.05$ *vs* VEH+SDS group (One-way ANOVA, followed by Bonferroni post-hoc test).

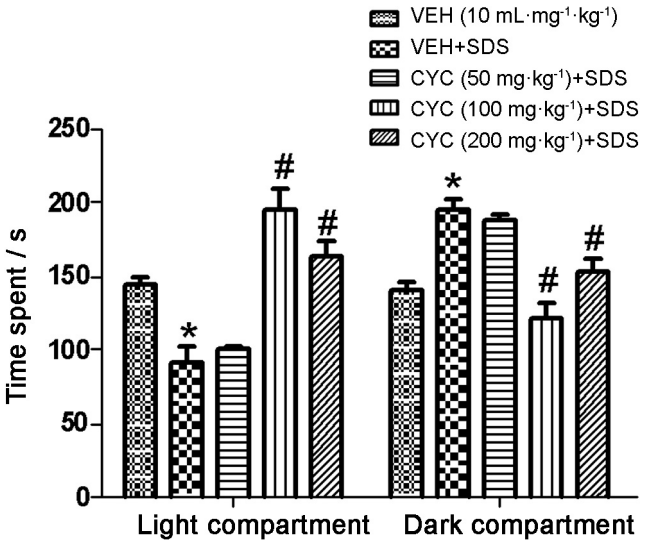

Fig. 2. Effect of aqueous leaf extract of *C. citratus* (CYC) on social defeat stress (SDS)-induced anxiety-like behavior in mice (mean ± S.E.M, n = 7). $^{*}P < 0.05$ *vs* vehicle (VEH) group; $^{\#}P < 0.05$ *vs* VEH+SDS group (One-way ANOVA, followed by Bonferroni post-hoc test).

3.3. C. citratus ameliorates depressive-like behavior in mice subjected to social defeat stress

As shown Fig. 3, One-way ANOVA and Post-hoc analysis revealed that SDS produced depressive-like behavior in mice as revealed by increased duration of immobility in the TST when compared with non-stress control. However, oral administration of aqueous leaf extract of *C. citratus* (50, 100 and 200 mg/kg, p.o.) significantly (F [4,30] = 27.26, $P < 0.0001$) reduced SDS-induced depressive-like effect.

3.4. C. citratus improves memory performance in socially defeated mice

The effect *C. citratus* on SDS-induced memory impairment was presented in Fig. 4. Mice exposed to chronic SDS had memory deficit in comparison with non-stress control as shown by decreased % alternations in the Y-maze test. However, *C. citratus* (50, 100 and

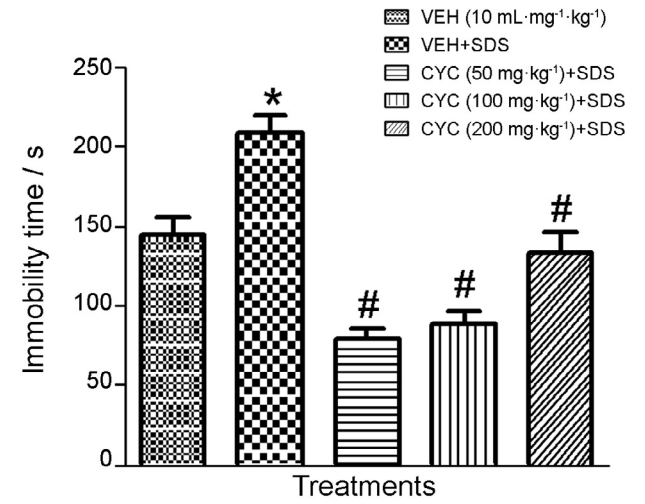

Fig. 3. Aqueous leaf extract of *C. citratus* (CYC) attenuates social defeat stress (SDS)-induced depressive-like behavior in mice (mean ± S.E.M, n = 7). $^{*}P < 0.05$ *vs* vehicle (VEH) group; $^{\#}P < 0.05$ *vs* VEH+SDS group (One-way ANOVA, followed by Bonferroni post-hoc test).

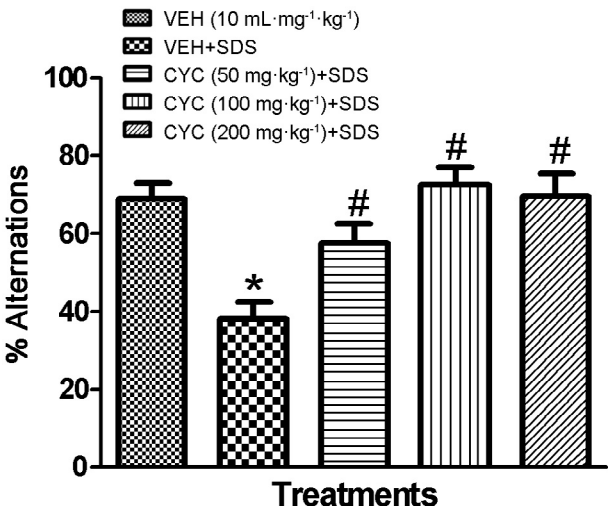

Fig. 4. Aqueous leaf extract of *C. citratus* (CYC) improves memory deficit in mice exposed to social defeat stress (SDS) (mean ± S.E.M, n = 7). $^{*}P < 0.05$ *vs* vehicle (VEH) group; $^{\#}P < 0.05$ *vs* VEH+SDS group (One-way ANOVA, followed by Bonferroni post-hoc test).

200 mg/kg, p.o.) significantly (F [4,30] = 9.203, $P < 0.0001$) attenuated the effect of SDS on memory performance in the Y-maze test.

3.5. C. citratus extract enhances social preference in socially defeat mice

The effect of *C. citratus* extract on the social preference of socially defeated mice was shown in Fig. 5. Mice exposed to SDS exhibited social avoidance in the social interaction test in comparison with control ($P < 0.05$). However, *C. citratus* extract (50, 100, and 200 mg/kg, p.o.) significantly (F [4,30] = 26.02, $P < 0.0001$) attenuated the deficit in social preference relative to SDS group.

3.6. C. citratus reduces oxidative stress in mice exposed to chronic social defeat stress

The effects of *C. citratus* on the concentrations of GSH and MDA in brains of mice exposed to SDS are shown in Figs. 6 and 7. As presented in Figs. 6 and 7, SDS produced marked increase in MDA and

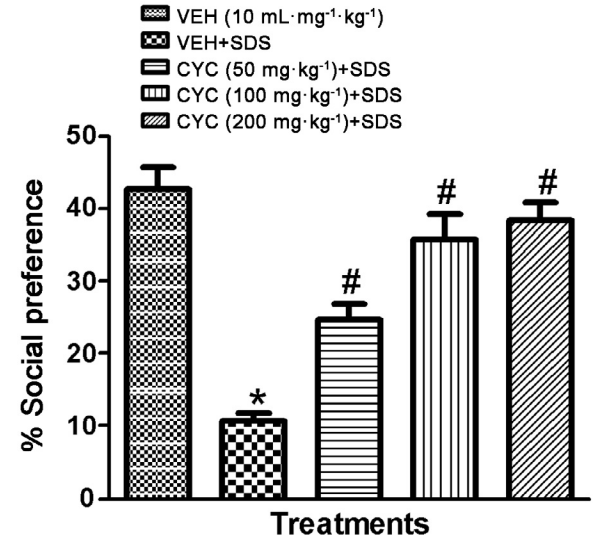

Fig. 5. *C. citratus* (CYC) aqueous leaf extract enhances social preference in socially defeat mice (mean ± S.E.M, n = 7). $^{*}P < 0.05$ *vs* vehicle (VEH) group; $^{\#}P < 0.05$ *vs* VEH +SDS group (One-way ANOVA, followed by Bonferroni post-hoc test).

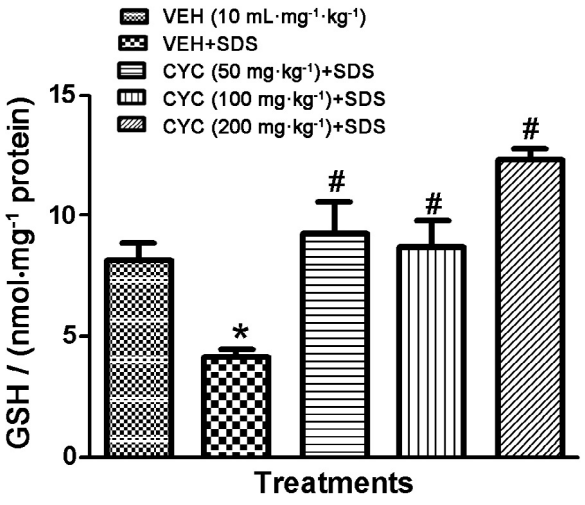

Fig. 6. *C. citratus* (CYC) leaf extract ameliorates social defeat stress (SDS)-induced depletion of brain glutathione (GSH) concentration in mice (mean ± S.E.M, n = 7). $^{*}P < 0.05$ *vs* vehicle (VEH) group; $^{\#}P < 0.05$ *vs* VEH+SDS group (One-way ANOVA, followed by Bonferroni post-hoc test).

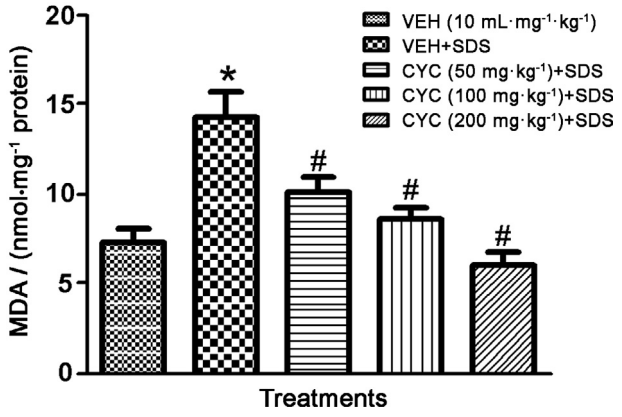

Fig. 7. Aqueous leaf extract of *C. citratus* (CYC) reduces social defeat stress (SDS)-induced increased brain malondialdehyde (MDA) contents in mice (mean ± S.E.M, n = 7). $^{*}P < 0.05$ *vs* vehicle (VEH) group; $^{\#}P < 0.05$ *vs* VEH+SDS group (One-way ANOVA, followed by Bonferroni post-hoc test).

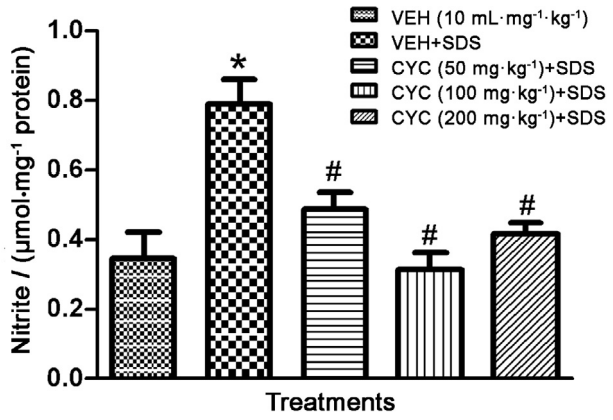

Fig. 8. *C. citratus* (CYC) leaf extract attenuates chronic social defeat stress-induced increase in nitrite concentration in mice (mean ± S.E.M, n = 7). $^{*}P < 0.05$ *vs* vehicle (VEH) group; $^{\#}P < 0.05$ *vs* VEH+SDS group (One-way ANOVA, followed by Bonferroni post-hoc test).

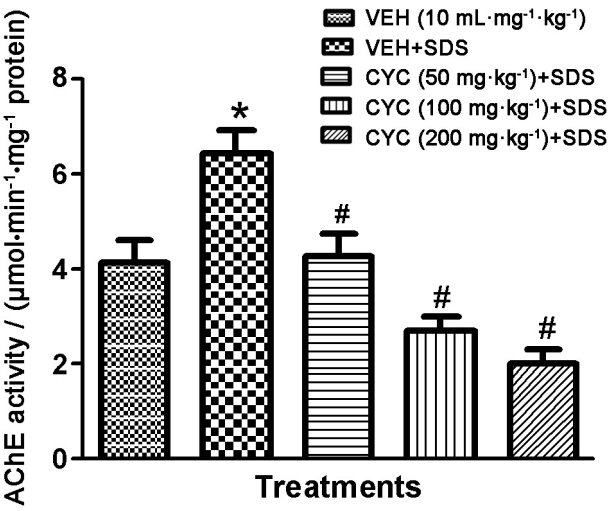

Fig. 9. *C. citratus* (CYC) aqueous leaf extract inhibits social defeat stress (SDS)-induced increased acetyl-cholinesterase (AChE) activity in mice brains (mean ± S.E.M, n = 7). $^{*}P < 0.05$ *vs* vehicle (VEH) group; $^{\#}P < 0.05$ *vs* VEH+SDS group (One-way ANOVA, followed by Bonferroni post-hoc test).

decreased GSH contents when compared with non-stress control. However, treatment with *C. citratus* (50, 100 and 200 mg/kg, p.o.) significantly (P < 0.05) reduced oxidative stress as shown by reduced MDA and increased GSH contents in the brain of mice exposed to SDS.

3.7. C. citratus reduces brain concentrations of nitrite and acetyl-cholinesterase activity in mice exposed to chronic social defeat stress

As presented in Fig. 8, mice exposed to SDS had increased brain contents of nitrite, which was significantly reversed by oral administration of *C. citratus* (50–200 mg/kg) (F [4,30] = 10.58, P < 0.05). SDS-induced the increase in AChE activity (Fig. 9) in mice brains was also attenuated by C. citratus (50–200 mg/kg) when compared with SDS control group (F [4,30] = 17.21, P < 0.0001).

4. Discussion

The results of this study revealed that *C. citratus* reverses SDS-induced hypolocomotion as indicated by increased SMA in defeated mice exposed to aggressive counterparts. SDS has been shown to produce multiple psychiatric symptoms in a defeated animal in the resident/intruder paradigm (Björkqvist, 2001; Lio et al., 2012; Solomon, 2017). Studies on social defeat in humans mostly focused on bullying in schools and in workplaces. Victims of bullying are known to suffer from depression, anxiety, phobia and loss of self-esteem, psychosomatic diseases, and other behavioral symptoms (Björkqvist, 2001; Solomon, 2017). Impaired locomotion is one of the prominent features experienced by animals exposed to uncontrollable chronic stress in various stress models including SDS (Gaurav et al., 2013). Besides, in clinical settings, the extent of motor activity is often used to indicate the severity of many chronic diseases as well as therapeutic benefits (Bennabi, Vandel, Pozzo, & Haffen, 2013). It is well reported in literature that impaired motor function affects the social interaction of animals and thus contributes to poor quality of life of patients recovering from post-traumatic stress or stressful life events (Venzala, García-García, Elizalde, Delagrange, & Tordera, 2012; Gaurav et al., 2013). The results of our study further confirmed that animals exposed to SDS present with impaired SMA in the open field test (Venzala et al., 2012). Thus, the ability of *C. citratus* to attenuate SDS-induced hypolocomotion in mice suggests that it

might mitigate the multiple adverse effects of chronic psychosocial stress on psychomotor functions in clinical settings.

Anxiety is a common neurobehavioral abnormality seen in animals exposed to chronic stress and a major feature of depressive disorder (Malick, 2010; Venzala et al., 2012). Our results showed that socially defeated mice exhibited heightened anxiety-like behavior as evidenced by increased duration of time spent in the dark compartment of the LDB. These results agree with previous findings showing the appearance of increased anxiety or anxious states in animals exposed to SDS in various animal models of anxiety (Krishnan et al., 2007; Venzala et al., 2012). However, anxiolytic agents have been shown to reverse SDS-induced freezing behavior, a condition akin to anxiety (Venzala et al., 2012). Our findings match well with previous studies, which have reported that *C. citratus* has anxiolytic or sedative activity in rodents (Costa et al., 2011). It is likely that this effect might have contributed to the ability of *C. citratus* to reduce the natural fearful reaction and apprehension associated with confrontation with a dominant organism.

Depression is a major neurological disorder with co-morbidity with other neuropsychiatric diseases (Carlini et al., 1986) hence; the test for depressive-like behavior using the TST was carried out in this study. Our finding further confirmed earlier investigations showing that SDS caused depressive-like behavior in rodents (Venzala et al., 2012). The usefulness of TST in the assessment of depressive-like behavior is based on the changes in the duration of immobility, which is commonly regarded as an index of 'behavioral despair' (Steru et al., 1985). Compounds that shortened the duration of immobility are judged to have antidepressant-like activity in this test (Steru et al., 1985). Previous studies have established that antidepressant drugs attenuated depressive-like symptom in rodents subjected to chronic SDS (Venzala et al., 2012. The results of this present study revealed that *C. citratus* reverses the increased immobility time in mice exposed to SDS, which suggest a potential benefit in alleviation of depressive-like behaviors associated with chronic stress. Meanwhile, several studies have shown that the crude extract and essential oils of lemongrass demonstrated antidepressant activity in rodents (Umukoro et al., 2017; Dudhgaonkar, 2014). Besides, various bioactive compounds such as luteolin, isoscoparin, quercetin, kaempferol and apigenin found in *C. citratus* (Figueirinha et al., 2008; Yang et al., 2009) might be playing a role in its ability to improve depressive-like symptom in mice exposed to SDS. However, further studies on these compounds are necessary to clarify this speculation.

Social avoidance is a neuropsychiatric phenomenon common to different disorders in humans, such as depression (Krishnan et al., 2007; Lagace et al., 2010; Venzala et al., 2012). Preclinical studies have shown that socially defeated mice by conspecific counterpart become persistently aversive toward social stimuli (Lagace et al., 2010). Most importantly, the finding that antidepressant treatments reversed SDS-induced social avoidance suggests that this behavioral pathology may be relevant to human depression (Lagace et al., 2010). Thus our finding is in agreement with previous investigation which showed that SDS induces social withdrawal behavior (Lagace et al., 2010). However, the ability of *C. citratus* to reduce social withdrawal behavior further indicates its usefulness in amelioration of depression in mice exposed to SDS. Experimental studies have shown that confrontations and subsequent subordination of an animal by a conspecific counterpart cause memory deficits in the defeated animals (Lagace et al., 2010; Lio et al., 2012; Gaurav et al., 2013). Thus, SDS is increasingly becoming popular as an animal model for evaluation of the deleterious effects of psychosocial stress on mental health (Lagace et al., 2010; Venzala et al., 2012). In this study, SDS impaired memory performance in the YMT in mice. These findings further support earlier studies, which have shown that SDS caused cognitive dysfunctions in rodents (Lagace et al., 2010; Umukoro, Kalejaye, Ben-Azu, & Ajayi, 2018). Our results revealed that *C. citratus* attenuates memory deficit in socially defeated mice, which further confirmed its potential benefit in conditions associated with cognitive decline (Lio et al., 2012).

Although multiple molecular and cellular signaling pathways orchestrate brain damage cause by chronic stress (Magarinos & McEwen, 1995; Malick, 2010), reactive oxygen and nitrogen species have been implicated in the death of neuronal cells in the brains of rodents exposed to chronic stress (Munhoz et al., 2008). Clinical studies have also confirmed the role of altered oxidative and nitrergic pathways in stress-induced neuropathology (Kanarik et al., 2001; Serrano & Klann, 2004). Increased levels of malondialdehyde, a major biomarker of oxidative stress and low levels of endogenous antioxidants have been found in chronic stress-induced behavioral deranged patients (Serrano & Klann, 2004). It is well known that increased oxidative stress and release of inflammatory mediators are major events involved in brain damage that often leads to vicious cycles for progressive neurodegeneration especially cholinergic pathways (Gaurav et al., 2013; Mineur et al., 2013). Previous studies have shown that loss of cholinergic neurons play crucial role in memory deficit induced by chronic stress (Klinkenberg & Blokland, 2010). In addition, both clinical and anatomical features such as altered synaptic plasticity, dendritic morphology and impaired neurogenesis have been described in several brain structures in chronically depressed patients and animals exposed to SDS (Iniguez et al., 2016). Our findings further confirmed previous investigations which revealed that SDS decreased antioxidant systems and increased biomarkers of oxidative/nitrergic stresses in rodents (Gaurav et al., 2013; Warren et al., 2013). Our results also support earlier investigations showing that the memory deficit caused by SDS was accompanied by increased brain acetylcholinesterase activity (Munhoz et al., 2008). Thus, inhibition of oxidative and nitrergic stresses is being proposed as a novel therapeutic target that could attenuate neuronal damage in social defeat stress-induced psychopathologies (Gaurav et al., 2013; Warren et al., 2013). Relevant to this, there are increasing lines of evidences that support the potentials of antioxidant supplementation in the prevention and treatment of stress-related diseases like depression and cognitive dysfunction (Gaurav et al., 2013; Panossian, 2013). Thus, inhibition of oxidative/nitrergic stresses and AChE activity may contribute to the ability of *C. citratus* in ameliorating depressive symptom and memory decline in mice exposed to SDS.

5. Conclusion

The results of this study provide evidences, which suggest that lemongrass may ameliorate the neurobiological features of various neuropsychiatric disorders in individuals exposed to chronic psychosocial stress or recovery from post-traumatic stress disorder.

Declaration of competing interest

There is no conflict of interest to declare.

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