Antidepressant-like Property of Jobelyn®, an African Unique Herbal Formulation in Mice

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Key words
- Jobelyn®
- forced swim test
- tail suspension test
- antidepressant

Abstract

Objectives: The purpose of this investigation was to evaluate whether Jobelyn® (JB) possesses anti-depressant-like property in the mouse forced swimming test (FST), tail suspension test (TST) and yohimbine-induced lethality test (YLT) in aggregated mice.

Methods: Mice were given JB (10–100 mg/kg, p.o.) daily for 7 days and then subjected to FST, TST, YLT and open field test. The parameters assessed in both FST and TST were the time (s) spent in active movement (struggling time), first occurrence of immobility (s) and the duration of immobility (s). In the YLT, the mortality rate was recorded 24 h after yohimbine (35 mg/kg, i.p.) administration. In the open field test, the number of line crosses and total distance travelled (m) were measured for 10 min in the open field chamber.

Results: JB significantly ($p < 0.05$) decrease the duration of immobility both in the FST and TST, which suggests antidepressant-like property. JB significantly ($p < 0.05$) prolonged the time spent in active swimming and delayed the first occurrence of immobility, indicating endurance promoting effect. It potentiated the toxic effect of yohimbine, which further suggests antidepressant-like activity and facilitation of both serotonergic and noradrenergic neurotransmissions. However, JB did not significantly increase the locomotor activity in the open-field test.

Conclusions: Jobelyn® has antidepressant-like activity, which may be related to the stimulation of serotonergic and noradrenergic pathways. The ability of Jobelyn® to delay the onset of immobility and to prolong the struggling time support its use as energizer in general body weakness or exhaustion.

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Introduction

Jobelyn® (JB) is a dietary supplement that has won both local and international recognition as anti-anaemic, immune booster and energizer [1]. JB is one of the fastest selling herbal medicines in Nigeria with most pharmaceutical outlets being used as the distribution channels. It is available as a powdered preparation formulated into capsules and suspensions for the treatment of moderate to severe anaemia (as in sickle cell patients), cancer and HIV/AIDS [2]. It is widely used to combat stress and to restore the much needed energy during the periods of recovery from debilitating diseases or sudden weight loss [1,2]. It is also claimed to be helpful in arthritis and neurological disorders [1–3]. Additionally, JB is also known to strengthen the immune system, thereby enhances body's defensive mechanisms in response to stress or pathogenic invasions [1,2]. The recommended dose by the manufacturer is 1 or 2 capsules (1–3 times daily) with each capsules consisting of 250 mg.

The active ingredients in JB were obtained from the leaves of a potent antioxidant Sorghum bicolor (Gramineae) plant that has been used for over a century to treat several diseases in African traditional medicine [1,2]. The common names of *Sorghum bicolor* include millet, guinea corn, broom corn, sweet sorghum and is cultivated principally for its nutritional and medicinal values [4]. The major active ingredients in JB include proanthocyanidins, anthocyanidins, apigenidins, proapigenidins, apigenins, luteolins and naringenins [1,2]. Most of these biologically active compounds have been found to exhibit a wide range of pharmacological activities [4,5]. In particular, apigenin, luteolin and naringenin have been shown to exhibit neuroprotection and to reduce neuroinflammation, which indicate their

therapeutic usefulness in central nervous system disorders [4–7]. Although previous studies have confirmed the anti-anaemic effect of JB [1,2], several other medicinal claims especially in central nervous system disorders are yet to be verified scientifically. This present study which describes the antidepressant property of JB is a part of our ongoing investigations on the psychopharmacological activities of this unique herbal formulation in experimental animals.

It is well recognized that a high levels of sustained stress is associated with the natural course of several illnesses especially depression [8]. Prolonged intense stress produced lowered mood and depletion of monoamines that played crucial role in the regulation of emotion and behavior [8,9]. Thus, a common neurochemical abnormality particularly noradrenergic system/serotonergic system exist between depressions and prolonged psychosocial stress [8–10]. Furthermore, depressed individuals and those under prolonged intense stress exhibit low tolerance to frustration and are more prone to exhaustion [8,11]. The use of JB as an energizer in the state of exhaustion and relieve of stress suggest that it might contain psychopharmacologically active compounds with anti-depressant property. Thus, this present investigation was carried out to evaluate whether Jobelyn® has anti-depressant-like property in rodent models predictive of endogenous depression in humans.

Materials and Methods

Experimental animals

Male albino Swiss mice (20–22 g) were obtained from the Central Animal House, University of Ibadan. The animals were housed in plastic cages at room temperature and they had free access to commercial food pellets and water ad libitum. They were acclimatized for at least one week before use for all experiments. The study was carried out in accordance with the ethical guidelines of the University of Ibadan for the care and use of laboratory animals for experimental studies.

Drugs and treatment

Imipramine (IM) hydrochloride (Sigma-Aldrich, St. Louis, USA), yohimbine (Sigma, USA) and Jobelyn® (Health Forever Products Ltd, Lagos, Nigeria) were used in the study. All drugs were dissolved in distilled water immediately before use and were given orally. The doses of 10, 50 and 100 mg/kg of JB used in the study were selected based on the results obtained from preliminary investigations. All the experimental procedures were started on day 7, 60 min after treatment.

Experimental Procedures

Forced swim test (FST)

The forced swim test is the most widely used test to evaluate depression-like behavior exhibited by mice [12]. The FST was carried out according to the method of Porsolt et al. [13]. Mice (6/group) were forced to swim individually in a glass jar (height: 20 cm, diameter: 10 cm) filled with water (depth: 15 cm) at a temperature of 25 ± 2 °C for 6 min. The duration of immobility (s) was recorded during the last 4 min of a 6 min observation period. A mouse was judged to be immobile when it remained floating in an upright position with the head above the water level. In addition, the struggling time (s) and first occurrence of immobility (s) were also measured. Struggling time was defined as the total time spent swimming with active limb motions during the 6 min test. Latency of immobility was defined as the duration from the start of the experiment to the first appearance of 4-limb immobility.

Tail suspension test (TST)

The TST was carried out according to the method previously described [14–16]. Mice (6/group) were individually suspended by the tail to a cord of about 50 cm in length stretched between 2 metal retort-stands at a height of 70 cm. After the initial 2 min period of vigorous motor activity, the mice became still and the immobility time (s) was measured with a stopwatch for a period of 4 min. Mice were considered immobile when they hung passively and completely motionless [14]. In addition, the struggling time (s) and first occurrence of immobility (s) were also measured. Struggling time was defined as the total time spent with active limb movements during the 6 min test. Latency of immobility was defined as the duration from the start of the experiment to the first appearance of 4-limb immobility.

Yohimbine lethality test

The antidepressant effect of JB was further evaluated utilizing the potentiation of yohimbine-induced lethality test in mice, as previously described [17]. Mice (10/group) received JB, 60 min before i.p. injection of yohimbine (35 mg/kg) and were immediately placed in cages. The numbers of death in each group were recorded 24 h after yohimbine administration.

Effect of Jobelyn on spontaneous motor activity (SMA)

The open field test was employed to screen the effect of JB on SMA in mice. Mice (6 per group) were given JB (10–100 mg/kg, p.o.), IM (25 mg/kg, p.o.), or vehicle (10 ml/kg, p.o.) 60 min before each animal was placed in the center of an open field chamber (72 cm × 72 cm × 36 cm). The number of line crosses and total distance travelled (m) for 10 min were recorded [18].

Statistical analysis

The data were expressed as mean ± S.E.M. The data were analyzed with Graph Pad Prism software version 4.03. Statistical analysis of data was done by One-way ANOVA, followed by Tukey post-hoc test. A level of $p < 0.05$ was considered as statistically significant.

Results

Effects of Jobelyn® on the performance of mice in the forced swim test

The effect of JB on performance of mice in the forced swim test, as measured by the first occurrence of immobility, duration of immobility and struggling time are presented in **Table 1**. One-way ANOVA showed that there were significant differences between treatment groups: first occurrence of immobility [$F(10, 55) = 77.14$, $p \leq 0.05$], duration of immobility [$F(10, 55) = 142.9$, $p < 0.05$] and struggling time [$F(10, 55) = 112.5$, $p < 0.05$]. Post-hoc analysis showed that JB (10, 50 and 100 mg/kg, p.o.) produced a significant decrease in the period of immobility when compared with control. Furthermore, JB significantly prolonged the struggling time and also delayed the first occurrence of immobility in comparison with control (**Table 1**).

Table 1 Effect of Jobelyn® on performance of mice in the forced swimming test.

Treatment	Struggling time (s)	First occurrence of immobility (s)	Duration of immobility (s)
Control	175.50 ± 5.25	71.00 ± 3.44	154.00 ± 5.27
JB 100 mg/kg	210.70 ± 4.09*	89.17 ± 3.41*	129.70 ± 2.86*
JB 50 mg/kg	282.20 ± 6.20*	97.67 ± 2.46*	58.17 ± 2.30*
JB 10 mg/kg	305.50 ± 4.11*	108.50 ± 3.05*	30.00 ± 3.53*
IM 25 mg/kg	326.20 ± 6.66*	154.20 ± 2.76*	24.50 ± 4.37*

Values represent the mean ± S.E.M for 6 animals per group. *$p < 0.05$ compared to control group (ANOVA followed by Tukey's post-hoc test)

Table 2 Effect of Jobelyn® on performance of mice in the tail suspension test.

Treatment	Struggling time (s)	First occurrence of immobility (s)	Duration of immobility (s)
Control	273.30 ± 3.94	72.67 ± 1.31	99.50 ± 2.46
JB 100 mg/kg	296.20 ± 5.26*	92.50 ± 1.46*	83.67 ± 1.54*
JB 50 mg/kg	304.80 ± 6.06**	94.83 ± 2.89*	65.83 ± 1.30*
JB 10 mg/kg	327.00 ± 3.39*	103.00 ± 3.82*	47.33 ± 1.93*
IM 25 mg/kg	356.30 ± 2.39*	242.50 ± 3.76*	43.50 ± 1.46*

Values represent the mean ± S.E.M for 6 animals per group. *$p < 0.05$ compared to vehicle control group (ANOVA followed by Tukey's post-hoc test)

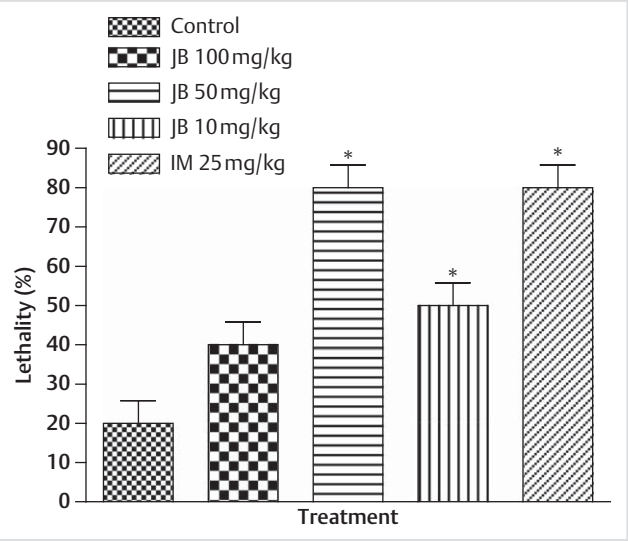

Fig. 1 Effect of Jobelyn® on yohimbine-induced lethality in aggregated mice. Each column represents the mean ± S.E.M (n = 10 per group). *$p < 0.05$ compared to control group (ANOVA).

Effects of Jobelyn® on the performance of mice in the tail suspension test

Table 2 showed the effects of JB on tail suspension test, as measured by first occurrence of immobility, duration of immobility and struggling time in mice. One-way ANOVA reveals that there were significant differences between treatment groups: first occurrence of immobility [$F(10, 55) = 281.3$, $p < 0.05$], duration of immobility [$F(10, 55) = 390.8$, $p < 0.05$] and struggling time [$F(10, 55) = 196.7$, $p < 0.05$]. Post-hoc analysis showed that the JB (10, 50 and100 mg/kg) and IM treated groups were significantly different ($p < 0.05$) when compared with control. JB significantly ($p < 0.05$) shortened the duration of immobility in the tail suspension test, which indicates antidepressant effect.

Table 3 Effect of Jobelyn® on spontaneous motor activity in mice.

Treatment	Number of line crosses	Total distance travelled (m)
Control	167.20 ± 4.73	3.01 ± 0.08
JB 100 mg/kg	165.80 ± 3.66	2.99 ± 0.07
JB 50 mg/kg	148.70 ± 3.69	2.68 ± 0.07
JB 10 mg/kg	169.30 ± 4.55	3.05 ± 0.08
IM 25 mg/kg	172.00 ± 3.10	33.10 ± 0.06

Values represent the mean ± S.E.M for 6 animals per group. JB did not significantly alter the locomotor activity when compared to vehicle control group ($p > 0.05$, ANOVA followed by Tukey's post-hoc test)

Jobelyn® potentiates lethality induced by yohimbine in aggregated mice

The effect of JB (10, 50 and 100 mg/kg, p.o.) on yohimbine-induced lethality are depicted in **Fig. 1**. Post-hoc analysis showed that JB (10–50 mg/kg, p.o.) significantly ($p < 0.05$) potentiated the lethal effect of yohimbine (35 mg/kg, i.p.), which was comparable to that of IM. However, at a dose of 100 mg/kg, JB did not significantly ($p > 0.05$) potentiate the toxic effect of yohimbine in mice (**Fig. 1**). Although, JB (10 mg/kg, 50 mg/kg) significantly potentiated the lethal effect of yohimbine, it did not produce any toxic symptoms or death in mice when given alone.

Effect of Jobelyn® on spontaneous motor activity

Table 3 indicates the effects of JB on locomotor activity in the open field test, as measured by the number of line crosses and total distance travelled (m). One-way ANOVA indicated that there were no significant differences in motor activity of the animals when treated with daily doses of JB (10–100 mg/kg, p.o.) for 7 days as compared with control: number of line crosses [$F(10, 55) = 4.11$, $p > 0.05$] and total distance travelled [$F(10, 55) = 4.14$, $p > 0.05$].

Discussion

The results of this study revealed that JB reduced the duration of immobility in the forced swim and tail suspension tests in mice, indicating antidepressant-like effect. JB also increase the ability of the animals to copy with the aversive situations, as shown by increase in struggling time and delay in the first occurrence of immobility in the FST and TST. The FST and TST are stress models widely used as valid behavioral paradigms for the evaluation of antidepressant drugs in rodents [13,14]. In both tests, rodents are exposed to an aversive situation from which there is no escape, and will, after periods of agitation, cease attempts to escape and become immobile [13,14]. Thus, the appearance of immobility indicates a state of exhaustion, lowered mood (hopelessness) or despairs and antidepressant drugs are known to decrease the period of immobility in rodents [12,19]. However, the anti-immobility effect of JB was not associated with central nervous system stimulation, as it did not significantly increase the SMA in the open-field test. This further suggests that the anti-immobility activity exhibited by JB reflects a true antidepressant-like effect. The YLT was further used to validate the antidepressant-like property of JB. The potentiation of yohimbine-induced lethality in aggregated mice has served as an additional paradigm for the routine screening of compounds with antidepressant activity [17]. Thus, the findings that JB enhanced the lethal effect of yohimbine further suggest that it may have antidepressant-like property.

The pathological basis of depression is closely linked with reduced brain levels of monoamines especially norepinephrine and serotonin [8,9]. Although the factors that trigger the depletion of these brain transmitters are not yet clearly established, prolonged intense stress appears to play a predominant role [8,9]. Both preclinical and clinical studies have confirmed that stress may precipitate depressive illness, as a consequence of the depletion of brain monoamines [19–21]. Thus, the most common models predictive of antidepressant activity are performed under intense stressful conditions in rodents [12]. In addition, antidepressant drugs are known to reduce the duration of immobility through facilitation of both serotonergic and noradrenergic neurotransmissions in these animal models [23–25]. Although further studies are necessary before drawing any conclusions on how JB exerts its antidepressant-like action, the data obtained from this present investigation suggests that it possesses phytochemicals that might be interacting with serotonergic and noradrenergic pathways. This suggestion is in accordance with the results obtained from the YLT, as yohimbine is known to interfere with both noradrenaline and serotonin functions [26,27]. Specifically, yohimbine induces lethality in grouped mice through the release of catecholamines from the brain and adrenal glands as consequence of antagonism of presynaptic α_2-adrenergic receptors [28,29]. In addition, antagonism of presynaptic α_2-adrenergic receptors promotes the release of serotonin, which further contributes to the overall toxicity caused by yohimbine [28,30]. Thus, the major event involved in yohimbine toxicity is an overall increase in serotonergic and noradrenergic neurotransmissions [17]. Compounds with antidepressant property are known to potentiate the lethal effect of yohimbine by enabling the amines to reach the receptors in greater amounts, either by inhibiting their reuptake or by reducing their inactivation [17,28,30]. Although the mode of interaction of JB with yohimbine in aggregated mice is yet to be established, previous studies had shown that naringenin, a phenolic compound presence in JB inhibited monoamine oxidase (MAO), an enzyme responsible for the inactivation of monoamines [5,6]. Thus, it is suggestive that the presence of naringenin in JB might be contributing to the overall toxicity of yohimbine in aggregated mice through the elevation of noradrenaline and serotonin levels. Furthermore, studies had also established the antidepressant activity of naringenin, as a consequence of MAO inhibition [5,6]. Therefore, it is likely that the presence of naringenin in JB might be playing a significant role in its antidepressant-like property observed in this study.

Psychomotor stimulants which are clinically ineffective as antidepressants are known to show antidepressant-like effects in the FST and TST [18]. Thus, to discount the possibility of false positives, JB was evaluated for its effects on SMA in the open field test. JB did not increase SMA in mice, which suggest that the anti-immobility effect, reflect a true antidepressant-like action, as antidepressant drugs are known to reduce immobility period at doses that do not cause a significant increase in locomotor activity of rodents in the open field tests [25].

Conclusion

The present study reveals that JB has antidepressant-like activity and may play a role in the elevation of mood in depressed patients. Furthermore, the ability of JB to delay the onset of immobility and to prolong the struggling time may contribute to its usefulness as an energizer in general body weakness or exhaustion.

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Conflict of Interest

There are no conflicts of interest to declare.

References

1 *Okochi VI*, *Okpuzor J*, *Okubena MO et al.* The influence of African Herbal Formula on the haematological parameters of trypanosome infected rats. African Journal of Biotechnology 2003; 2: 312–316
2 *Erah PO*, *Asonye CC*, *Okhamafe AO*. Response of *Trypanosoma brucei brucei* – induced anaemia to a commercial herbal preparation. African Journal of Biotechnology 2003; 2: 307–311
3 *Oshikoya KA*, *Senbanjo IO*, *Njokanma OF et al.* Use of complementary and alternative medicines for children with chronic health conditions – in Lagos, Nigeria. BMC Complementary and Alternative Medicine 2008; 8: 66
4 *Awika JM*, *Rooney LW*. Sorghum phytochemicals and their potential impact on human health. Phytochemistry 2004; 65: 1199–1221
5 *Yi LT*, *Li CF*, *Zhan X et al.* Involvement of monoaminergic system in the antidepressant-like effect of the flavonoid naringenin in mice. Prog Neuropsychopharmacol Biol Psychiatry 2010; 34: 1223–1228
6 *Olsen HT*, *Stafford GI*, *Standen JV et al.* Isolation of the MAO-inhibitor naringenin from Mentha aquata L. Journal of Ethnopharmacology 2008; 117: 500–502
7 *Heo HJ*, *Kim M*, *Lee J et al.* Naringenin from *Citrus junos* has an inhibitory effect on acetylcholinesterase and a mitigating effect on amnesia. Dementia and Geratric Cognitive Disorders 2004; 17: 151–157
8 *Baars MY*, *Müller MJ*, *Gallhofer B et al.* Depressive and Aggressive Responses to Frustration: Development of a Questionnaire and Its Validation in a Sample of Male Alcoholics. Depression Research and Treatment 2011; 2011: 1–19
9 *VanPraag HM*. "Anxiety/aggression-driven depression". Progress in Neuro-Psychopharmacology and Biological Psychiatry 2001; 25: 893–924
10 *Åsberg M*, *Träskman L*, *Thoren P*. "5 HIAA in the cerebrospinal fluid. A biochemical suicide predictor?" Archives of General Psychiatry 1976; 33: 1193–1197
11 *Taki M*, *Tam FW*. Bullying among girls in Japan and Hong Kong: An examination of the frustration-aggression model. Educational Research and Evaluation 2007; 13: 373–399
12 *Roche M*, *Commons KG*, *Peoples A et al.* Circuitry underlying regulation of the serotonergic system by swim stress. J Neurosci 2003; 23: 970–977
13 *Porsolt RD*, *Anton G*, *Deniel M et al.* Behavioral despair in rats: a new animal model sensitive to antidepressant treatments. Eur J Pharmacol 1978; 47: 379–391
14 *Steru L*, *Chermat R*, *Thierry B et al.* The tail suspension test: a new method for screening antidepressants in mice. Psychopharmacology 1985; 85: 367–370
15 *Cryan JP*, *Mombereau C*, *Vassout A*. The tail suspension test as a model for assessing antidepressant activity: review of pharmacological and genetic studies in mice. Neurosci Biobehav Rev 2005; 29: 571–625
16 *Zomkowski AD*, *Santos AR*, *Rodrigues AL*. Putrescine produces antidepressant-like effects in the forced swimming test and in the tail suspension test in mice. Prog Neuropsychopharmacol Biol Psychiatry 2006; 30: 1419–1425
17 *Malick JB*. Potentiation of yohimbine-induced lethality in mice: predictor of antidepressant potential. Drug Dev Res 1983; 3: 357–363
18 *Sherman AD*, *Sacquitne JL*, *Petty F*. Specificity of the learned helplessness model of depression. Pharmacol Biochem Behav 1982; 16: 449–454
19 *Subarnas A*, *Tadano T*, *Nakahata N et al.* A possible mechanism of antidepressant activity of beta-amyrin palmitate isolated from *Lobelia inflata* leaves in the forced swimming test. Life Sciences 1993; 52: 289–296

20 *Blair RJR*. "Psychopathy, frustration, and reactive aggression: the role of ventromedial prefrontal cortex". British Journal of Psychology 2010; 101: 383–399
21 *Anguelova M*, *Benkelfat C*, *Turecki G*. A systematic review of association studies investigating genes coding for serotonin receptors and the serotonin transporter: II. Suicidal behavior. Mol Psychiatry 2003; 8: 646–653
22 *Van Praag HM*. Serotonin-related, anxiety/aggression-driven, stressor-precipitated depression. A psycho-biological hypothesis. European Psychiatry 1996; 11: 57–67
23 *Hajos KE*, *Mctavish SF*, *Sharp T*. Effect of a selective 5-hydroxytryptamine reuptake inhibitor on brain extracellular noradrenaline: microdialysis studies using paroxetine. Eur J Pharmacol 2000; 407: 101–107
24 *Adell A*, *Castro E*, *Celada P et al*. Strategies for producing faster acting antidepressants. Drug Discov Today 2005; 10: 578–585
25 *Kirby LG*, *Lucki I*. Interaction between the forced swimming test and fluoxetine treatment on extracellular 5-hydroxytryptamine and 5-hydroxyindoleacetic acid in the Rat. J Pharmacol Exp Ther 1997; 282: 967–976
26 *Papeschi R*, *Sourkes TL*, *Youdim MBH*. The effect of yohimbine on brain serotonin metabolism, motor behavior and body temperature of the rat. Eur J Pharmacol 1971; 15: 318–326
27 *Söderpalm A*, *Blomqvist O*, *Söderpalm B*. The yohimbine-induced anti-conflict effect in the rat. I: Involvement of noradrenergic, serotonergic and endozepinergic(?) mechanisms. Eur J Pharmacol 1971; 15: 318–326
28 *Quinton RM*. The increase in the toxicity of yohimbine induced by imipramine and other drugs in mice. Br J Pharmacol Chemother 1963; 21: 51–66
29 *Goldberg MR*, *Robertson D*. Yohimbine: a pharmacological probe for study of the α2-adrenoceptor. Pharmacol Rev 1983; 35: 143–180
30 *Siqueira IR*, *Lara FS*, *Silva DR et al*. Psychopharmacological properties of *Ptychopetalum olacoides* Bentham (Olacacaeae). Pharm Biol 1998; 36: 327–334

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