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Antidepressant effects of curcumin in the forced swim test and olfactory bulbectomy models of depression in rats

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Abstract

Curcuma longa is a major constituent of Xiaoyao-san, the traditional Chinese medicinal formula, which has been used to effectively manage stress and depression-related disorders in China. Curcumin is the active component of curcuma longa, and we hypothesized that curcumin would have an influence on depressive-like behaviors. The purpose of the present study was to confirm the putative antidepressant effect of chronic administrations of curcumin (1.25, 2.5, 5 and 10 mg/kg, p.o.) in the forced swimming test and bilateral olfactory bulbectomy (OB) models of depression in rats. In the first study, chronic treatment with curcumin (14 days) reduced the immobility time in the forced swimming test. In the second experiment, curcumin reversed the OB-induced behavioral abnormalities such as hyperactivity in the open field, as well as deficits in step-down passive avoidance. In addition, OB-induced low levels of serotonin (5-HT), noradrenaline (NA), high 5-hydroxyindoleacetic acid (5-HIAA) and 4-dihydroxyphenylacetic acid (DOPAC) in the hippocampus were observed, and were completely reversed by curcumin administration. A slight decrease in 5-HT, NA and dopamine (DA) levels was found in the frontal cortex of OB rats which was also reversed by curcumin treatment. These results confirm the antidepressant effects of curcumin in the forced swim and the OB models of depression in rats, and suggest that these antidepressant effects may be mediated by actions in the central monoaminergic neurotransmitter systems.

Keywords: Curcumin; Antidepressant; Forced swimming; Olfactory bulbectomy; Open field; Passive avoidance; Brain monoamines; Rats

1. Introduction

Depressive disorders are among the most frequently occurring psychiatric diseases with prevalence rates between 9% and 18% in the Western world (Schloss and Henm, 2004). Although the current pharmacotherapy of depression includes a battery of drugs, many are inconsistently effective and can exert undesirable side effects. The development of safe and powerful antidepressant agents from traditional herbs may therefore alleviate some of the side effects that accompany antidepressant drugs.

Many traditional Chinese medicines, such as Xiaoyao-san, have been used successfully to manage depressive disorders (Kong et al., 2001; Chen and Tang, 2004). Curcuma longa is commonly found in Chinese herbal medicines including Xiaoyao-san, which is used to promote the flow of liver *qi* and treat the symptoms of mental stress, hypochondriac distensive pain and mania. The antidepressant effects of curcuma longa have also been investigated with behavioral despair tests in mice (Yu et al., 2002). As the major constituent of curcuma longa, curcumin possesses many therapeutic properties including antioxidant, anti-inflammatory, immunodulatory and neuroprotective activities (Motterlini et al., 2000; Thiyagarajan and Sharma, 2004). However, information regarding the antidepressant activity of curcumin is lacking. Previous studies have shown that curcumin inhibits the activity of monoamine oxidase (MAO) in C6 glial cells; MAO plays a central role in several psychiatric neurological disorders, including clinical depression and anxiety (Mazzio et al., 1998). There is evidence that MAO inhibitor-induced increases in monoaminergic neurotransmission can alleviate clinical depression (Dar and Khatoon, 2000).

We assessed the antidepressant effect of curcumin in a pilot study and observed that acute administration of curcumin (2.5, 5 and 10 mg/kg, p.o.) significantly decreased the immobility time in the forced swim test and tail suspension test in mice,

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indicating a possible antidepressant effect (unpublished results). Based on these results, in the current study we assessed the antidepressant effects of chronically administered curcumin in the forced swim test and bilateral olfactory bulbectomy (OB) rat models of depression. In addition, because the central monoaminergic systems are believed to contribute to the etiology of depression, the concentrations of monoamines in the hippocampus and the frontal cortex of sham control and OB rats following chronic curcumin administration were examined.

2. Materials and methods

2.1. Animals

Male Sprague–Dawley rats (200–250 g) were obtained from the Department of Laboratory Animal Science, Peking University Health Science Center (Beijing, China). The animals were housed six per cage under standard colony conditions, with a 12 h light/dark cycle and ad libitum food and water. They were allowed to acclimatize to the colony for at least 7 days prior to any experimentation. The experiment procedures were in compliance with the National Institutes of Health Guide for Care and Use of Laboratory Animals and with the European Communities Council Directive of 24 November 1986 (86/609/EEC).

2.2. Drugs and drug administration

Curcumin, imipramine hydrochloride, 5-hydroxytryptamine (5-HT), noradrenaline (NA), dopamine (DA), 5-hydroxyindoleacetic acid (5-HIAA) and 4-dihydroxyphenylacetic acid (DOPAC) were purchased from Sigma Chemical Co., (USA). For oral (p.o.) administration, curcumin was dissolved in peanut oil and diluted to the desired concentration on the day of experiment. For intraperitoneal (i.p.) injection, imipramine was dissolved in double-distilled water. In this study, various doses of curcumin (1.25, 2.5, 5, 10 mg/kg) were administered (p.o.) and imipramine (10 mg/kg) was injected (i.p.) for 14 days. The experiments were conducted 60 min after the last drug treatment. In a preliminary experiment, peanut oil and redistilled water were used as control treatments and the behavioral data did not differ between the rats that received the two vehicle solutions. Therefore, we chose to present only the peanut oil control group data for comparison.

2.3. Forced swim test

On the 14th day of chronic curcumin administration, the forced swim test was performed. This study was carried out in rats ($n=8$/group) according to the methods described by Porsolt et al. (1978). Briefly, rats were placed individually in glass cylinders (height: 40 cm, diameter: 18 cm) containing 23 cm of water at 25 °C. Fifteen minutes later, rats were removed and dried before being returned to their home cages. The animals were replaced in the cylinders 24 h later, and the procedure was repeated, but on this occasion the duration that the rats remained immobile during a 5-min observation period was recorded.

2.4. Olfactory bulbectomy

After a week of acclimation to the vivarium, bilateral olfactory bulbectomy was performed in rats ($n=6–7$/group) anesthetized with 2.5% w/v tribromoethanol (10 mg/kg, i.p.) essentially as described by Nowak et al. (2003). Briefly, following exposure of the skull, holes were drilled 7 mm anterior to bregma and 2 mm on either side of the midline at a point corresponding to the posterior margin of the orbit of the eye. The olfactory bulbs were removed by suction, the holes were filled with haemostatic sponge in order to control the bleeding and the scalp was sutured. To prevent infection, the animals were given 40,000 IU/kg of procaine penicillin by intramuscularly injection after surgery. Sham-operated animals received the same surgical treatment, but the bulbs were left intact. The animals were given 14 days to recover following the surgery prior to drug administration, and they were handled daily by the experimenter throughout the recovery period to reduce stress and/or aggressive behavior.

2.5. Open field test

OB and sham control rats were subjected to an open field test on the 14th day of chronic curcumin administration. Each rat was placed individually into the center of the open field apparatus. This apparatus was essentially as described by Redmond et al. (1999) with a slight modification. The open field apparatus consisted of a 80-cm diameter arena with 75-cm high aluminum walls, divided into 10 cm^2. A 60 W light bulb was positioned 90 cm above the base of the arena, and provided the only source of illumination in the testing room. Each animal was placed in the center of the open field apparatus, and the ambulation scores (the number of squares crossed) and the number of rearings and peepings were measured during a 3-min period.

2.6. Passive avoidance test

OB and sham control rats were also subjected to step-down passive avoidance training and testing on the 14th day of chronic curcumin administration. The procedures were similar to those described by Pilc et al. (2002). The apparatus consisted of an open box ($50\times50\times50$ cm^3) with black walls and a stainless steel grid floor. The electrified rods were 1.2 cm apart and were connected to the terminals of a shock generator that delivered square wave pulses with a constant voltage. The delivered shock had constant intensity (0.75 mA) and lasted 1 s. A wooden platform ($12\times12\times4$ cm^3) was in the center of the box. Each rat was placed on this platform, and when it stepped off the platform with all four paws it received an electric shock. The animal was immediately removed from the experimental apparatus and placed in its home cage. After 30 s, the next trial was initiated. Each rat was trained until it learned to remain on the platform for 1 min or for 15 trials, whichever occurred first.

The total number of training trials required for the rat to reach the 1 min criterion was recorded as the learning index. Rats that did not learn to avoid the shock for ≥1 min were given the maximum score of 15.

2.7. Determination of monoamines and metabolites

Following behavioral testing, the sham-operated and OB-operated rats were decapitated and their brains were rapidly removed and frozen on dry ice. Brain regions containing the frontal cortex and hippocampus were dissected on a cold plate (−16 °C) (Franklin and Paxinos, 1997). The tissue samples were weighed and stored at −80 °C until homogenization.

Serotonin (5-HT), NA, DA, 5-HIAA and DOPAC levels in frontal cortex and hippocampus were measured as described previously (Nitta et al., 1992) using high-performance liquid chromatography (HPLC) with electrochemical detection, with minor modifications. Briefly each frozen tissue sample was homogenized by ultrasonication in 200 μl of 0.4 M perchloric acid (solution A). The homogenate was kept on ice for 1 h and then centrifuged at 12,000 × *g* (4 °C) for 20 min, and the pellet was discarded. An aliquot of 160 μl of supernatant was added to 80 μl of solution B (containing 0.2 M potassium citrate, 0.3 M di-potassium hydrogen phosphate and 0.2 M EDTA). The mixture was kept on ice for 1 h and then centrifuged at 12,000 × *g* (4 °C) for 20 min again. Twenty microliters of the resultant supernatant was directly injected into an ESA liquid chromatography system equipped with a reversed-phase C_{18} column (150 × 4.6 mm I.D., 5 μm) and an electrochemical detector (ESA CoulArray, Chelmstord, MA, USA). The detector potential was set at 50, 100, 200, 300, 400, 500 mV, respectively. The mobile phase consisted of 125 mM citric acid-sodium citrate (pH 4.3), 0.1 mM EDTA, 1.2 mM sodium octanesulfonate and 16% methanol; the flow rate was 1.0 ml/min. The tissue levels of monoamines were expressed in terms of nanograms per gram of tissue.

2.8. Statistical analyses

The data are presented as means ± standard errors of the means (S.E.M.). Differences were estimated by one-way analysis of variance (ANOVA) followed by Dunnet's *t*-test in the forced swim test. Data were analyzed using a two-way ANOVA where drug treatment and bulbectomy were the first and second factors in the OB test. Individual group differences were further assessed with the Fishers (least significant difference) LSD multiple range tests to determine the sources of any significant ANOVA comparisons. Differences with $P<0.05$ were considered statistically significant.

3. Results

3.1. The effects of curcumin in the forced swim task

Chronic curcumin administration reduced, in a dose-dependent manner, immobility time in the forced swim test (Fig. 1). Rats that were given daily curcumin doses of 1.25, 2.5, 5 or 10 mg/kg (p.o.) for 14 days exhibited 18.7%, 38.7%, 63.5% and 68.9% reduced immobility as compared with the control group, respectively. The effects of curcumin were similar to those observed for the classical antidepressant imipramine (10 mg/kg, i.p.). The percentage of inhibition for rats given imipramine was 60.6% in the forced swim test.

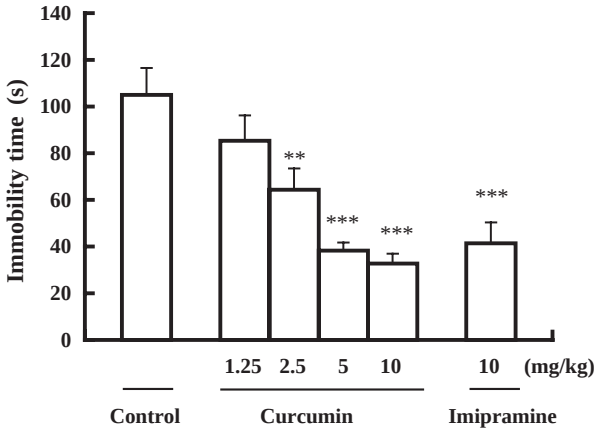

Fig. 1. The effects of chronic curcumin administration on the immobility time in the rat forced swimming test. Rats were given vehicle, curcumin (p.o., at doses of 1.25, 2.5, 5 and 10 mg/kg) or imipramine (10 mg/kg) for 14 days prior to testing. 60 min following the last treatment, the immobility time was recorded during a 5-min period of the swim sessions. The respective percent reduction in immobility time was 18.7%, 38.7%, 63.5% and 68.9% for curcumin at 1.25–10 mg/kg and it was 60.6% for imipramine at 10 mg/kg. Each value represents the mean ± S.E.M. of eight rats per group. $^{**}P<0.01$, $^{***}P<0.01$, compared with control group.

3.2. The effects of curcumin in the open field test in OB rats

In the open field test, an increase in activity (ambulation counts, rearings and peepings) was found in the OB group as compared with the sham-operated group. A two-way ANOVA revealed extremely significant effects of OB surgery [$F(1,56)=6.60$, $P<0.05$] and curcumin administration [$F(6,56)=15.91$, $P<0.001$] on ambulation, as well as a curcumin × OB interaction [$F(5,56)=2.83$, $P<0.05$]. Moreover, there was a significant attenuation of OB-related hyperactivity in the groups that were given 5 and 10 mg/kg curcumin. Two-way ANOVAs also revealed significant effects of OB [$F(1,56)=15.07$, $P<0.01$] and curcumin administration [$F(6,56)=6.67$, $P<0.01$] on the number of rearings and peepings, as well as a curcumin × OB interaction [$F(5,56)=3.98$, $P<0.01$]. Similarly, there was also a significant attenuation of OB-related hyperactivity in the 5 and 10 mg/kg curcumin and 10 mg/kg imipramine administration groups (Table 1).

3.3. The effects of curcumin on passive avoidance in OB rats

As shown in Fig. 2, sham-operated animals learned the passive avoidance task in approximately four trials. Rats with bilateral olfactory bulb ablation needed an average of eleven trials to reach criterion. A two-way ANOVA revealed a significant effect of OB surgery [$F(1,56)=32.16$, $P<0.01$] and curcumin administration [$F(6,56)=3.69$, $P<0.01$] on

Table 1
The effects of chronic curcumin administration on open field behavior in the olfactory bulbectomy model of depression in rats

Group	Ambulation counts	Number of rearings and peepings
Sham+vehicle	68.0±3.5	11.6±1.2
Sham+1.25 mg/kg curcumin	65.0±4.1	10.0±0.6
Sham+2.5 mg/kg curcumin	67.8±3.6	9.5±0.6
Sham+5 mg/kg curcumin	59.8±6.6	8.7±2.6
Sham+10 mg/kg curcumin	58.7±5.0	9.8±1.5
Sham+10 mg/kg imipramine	25.0±6.0###	6.8±1.6#
OB+vehicle	100.8±8.4##	21.8±2.1##
OB+1.25mg/kg curcumin	96.8±6.9	20.5±2.4
OB+2.5mg/kg curcumin	79.2±9.5	17.0±4.2
OB+5mg/kg curcumin	61.7±8.7**	13.3±2.1*
OB+10mg/kg curcumin	51.0±9.5***	8.5±1.2**
OB+10mg/kg imipramine	42.0±6.4***	6.4±1.2***

The open field test was performed 60 min after the last administration of chronic curcumin or imipramine treatment. Each column represents the mean±S.E.M. of six to seven rats per group.
# $P<0.05$, compared with sham+vehicle.
$P<0.01$, compared with sham+vehicle.
$P<0.001$, compared with sham+vehicle.
* $P<0.05$, compared with OB+vehicle.
** $P<0.01$, compared with OB+vehicle.
*** $P<0.001$, compared with OB+vehicle.

passive avoidance behavior, as well as a significant curcumin × OB interaction [$F(5,56)=2.94$, $P<0.05$]. Like imipramine (10 mg/kg), curcumin (5 and 10 mg/kg) administration attenuated the learning deficit in OB rats.

3.4. The effects of curcumin on monoamine and monoamine metabolite levels

3.4.1. Hippocampus

The levels of monoamines and monoamine metabolites detected in the hippocampus are summarized in Table 2. OB surgery [ANOVA, $F(1,56)=24.10$, $P<0.01$] and chronic curcumin treatment [ANOVA, $F(5,56)=4.01$, $P<0.01$] had significant effects on hippocampal 5-HT levels. Further analysis showed that OB surgery resulted in a decrease in the concentration of 5-HT measured, as compared with sham controls ($P<0.01$). Meanwhile, curcumin (5 and 10 mg/kg, p.o.) administration produced significant increases in 5-HT levels (P's<0.01) in the OB rats, but not in sham-operated rats.

A two-way ANOVA revealed a significant effect of OB surgery [$F(1,56)=14.87$, $P<0.01$], but not of the curcumin treatment [$F(5,56)=1.52$, $P>0.05$] on NA levels. The OB-operated vehicle-treated controls had a decreased mean NA concentration relative to sham-operated vehicle-treated controls ($P<0.05$). There was a trend toward increased NA levels with increased curcumin dose in the OB-treated groups, however, the effect was relatively small compared to the large NA deficit that resulted from OB surgery. A similar trend toward increased DA levels with increasing curcumin dose was observed in the OB rats. In addition, OB groups had increased levels of 5-HIAA and DOPAC (P's<0.01) relative to their respective sham control groups. Curcumin treatment at 10 mg/kg produced a significant decrease in 5-HIAA levels and a trend toward decreased DOPAC levels. Imipramine induced increases in 5-HT and NA levels in this region (Table 2).

3.4.2. Frontal cortex

The levels of monoamines and monoamine metabolites detected in the frontal cortex are summarized in Table 3. A two-way ANOVA showed a significant effect of OB surgery [$F(1,56)=4.82$, $P<0.05$] and a curcumin × OB interaction [$F(5,56)=2.77$, $P<0.05$] on 5-HT levels in the frontal cortex. Although no significant difference of curcumin treatment was found [$F(5,56)=1.19$, $P>0.05$], there was a trend towards decreased 5-HT levels in OB vehicle controls as compared with sham-operated rats. Moreover, curcumin (5 and 10 mg/kg) administration significantly elevated 5-HT levels in OB-operated animals as compared with the respective vehicle controls (P's<0.05). There were trends towards decreased NA and DA levels, and increased 5-HIAA and DOPAC levels, in OB controls relative to sham controls. Curcumin at

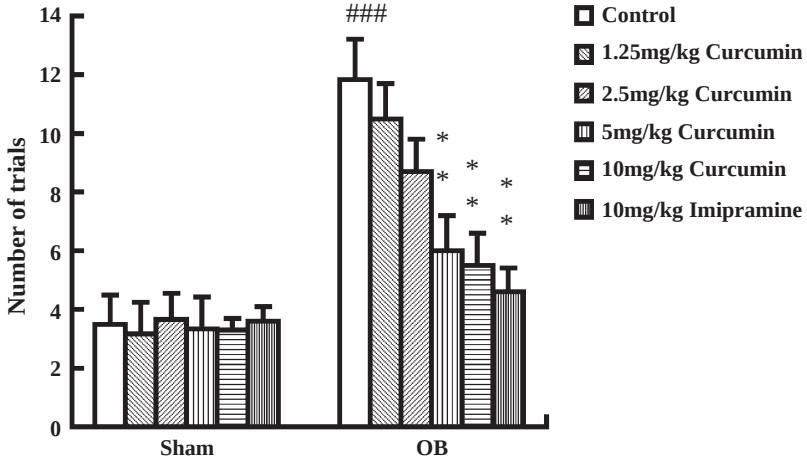

Fig. 2. The effects of chronic curcumin administration on passive avoidance acquisition in an olfactory bulbectomy model in rats. Passive avoidance training and testing was initiated 60 min after the last administration of chronic treatment with curcumin or imipramine. Each column represents the mean±S.E.M. from six to seven rats per group. ###$P<0.001$, compared with sham control. **$P<0.01$, compared with OB control.

Table 2
The effects of chronic curcumin administration on monoamine and monoamine metabolite levels in the hippocampus of OB and sham-operated rats

Group	5-HT	5-HIAA	NA	DA	DOPAC
Sham+vehicle	277.3±26.1	203.9±7.9	310.6±33.2	254.3±19.8	53.47±9.6
Sham+curcumin (1.25 mg/kg)	260.3±25.5	194.5±7.4	312.2±24.7	247.3±26.5	46.3±12.9
Sham+curcumin (2.5 mg/kg)	282.7±36.8	191.2±5.9	301.4±23.9	257.6±19.95	41.3±8.8
Sham+curcumin (5 mg/kg)	288.4±30.2	187.3±8.6	322.5±26.1	256.5±22.4	43.6±9.3
Sham+curcumin (10 mg/kg)	297.3±24.4	172.8±11.7	321.4±31.9	264.4±24.7	41.4±8.8
Sham+imipramine (10 mg/kg)	348.8±30.5	173.2±20.7	347.9±33.4	245.9±20.6	47.2±6.2
OB+vehicle	169.1±7.7##	285.3±27.9#	222.9±22.1#	232.2±22.1	87.6±13.5#
OB+curcumin (1.25 mg/kg)	191.9±25.5	259.2±43.1	224.9±27.7	240.9±24.4	73.8±15.4
OB+curcumin (2.5 mg/kg)	193.1±18.5	251.6±45.4	235.8±18.0	248.5±24.7	74.1±14.8
OB+curcumin (5 mg/kg)	245.8±20.6**	246.2±21.9	278.5±23.9	259.3±17.9	67.9±8.6
OB+curcumin (10 mg/kg)	260.5±17.6**	190.1±33.2*	288.5±24.9	276.9±18.9	61.9±9.9
OB+imipramine (10 mg/kg)	272.5±33.9**	209.4±18.4	297.5±24.5*	240.6±19.5	81.7±15.2

Data are expressed as means±S.E.M. in units of ng/g (n=6–7).
# P<0.05, compared with sham+vehicle.
P<0.01, compared with sham+vehicle.
* P<0.05, compared with OB+vehicle.
** P<0.01, compared with OB+vehicle.

10 mg/kg elevated NA and DA levels in the frontal cortex of OB-operated groups (P's<0.05 vs. vehicle-treated controls). Similarly, frontal cortex 5-HT and NA levels were increased in the OB rats (P's<0.01 vs. vehicle-treated controls) that were administered imipramine treatment (10 mg/kg, i.p.) (Table 3).

4. Discussion

In the present study, the antidepressant effects of curcumin were evaluated in two behavioral models, the forced swim task and the OB rat model of depression. Chronic administration of curcumin resulted in a dose-dependent reduction of the immobile time in the forced swim test. Curcumin treatment also reversed the increased activity in the open field test and the deficit in step-down passive avoidance learning deficit displayed by OB rats. These behavioral data are in agreement with previous work examining the effects of chronic treatment with both typical and atypical antidepressants (Janscar and Leonard, 1983).

As mentioned earlier, acute curcumin administration was previously shown to decrease the immobile time in the forced swim task, which is a behavioral paradigm that has been widely used as a screening assay for antidepressant activity (Vázquez-Palacios et al., 2004). In the present study, we demonstrated that chronic curcumin administration had an effect on immobility in the forced swim task that was similar to that seen with the acute treatment. Furthermore, we did not observe any evidence indicating the development of tolerance to the drug in rats that were given the drug for 14 consecutive days. In addition, preliminary studies showed that curcumin administered in doses effective in the forced swim test did not change the exploratory activity in the open field test. In fact, the results observed with curcumin treatment were largely comparable to those observed with the classical antidepressant drug imipramine. Therefore, our data suggest that curcumin produces selective antidepressant effects.

The mechanisms underlying the development of depression in OB rats are not known. However, these effects do not appear to simply be the result of anosmia as selective ablation of the

Table 3
The effects of chronic curcumin administration on monoamine and metabolite levels in the frontal cortex of OB and sham-operated rats

Group	5-HT	5-HIAA	NA	DA	DOPAC
Sham+vehicle	549.5±36.1	180.0±20.3	430.7±65.3	220.9±26.0	46.3±6.8
Sham+curcumin (1.25 mg/kg)	522.3±19.2	149.1±29.3	401.8±68.5	213.6±22.9	43.1±6.2
Sham+curcumin (2.5 mg/kg)	513.3±20.1	146.1±31.9	415.2±78.9	220.4±26.9	48.2±11.5
Sham+curcumin (5 mg/kg)	497.4±17.5	137.3±24.9	406.7±70.4	212.4±20.4	40.8±7.4
Sham+curcumin (10 mg/kg)	521.3±43.3	104.9±8.9#	482.5±52.7	222.6±22.3	46.6±9.2
Sham+imipramine (10 mg/kg)	507.4±14.8	85.5±9.1#	486.8±83.9	184.4±15.0	41.9±8.3
OB+vehicle	473.1±29.1	216.9±11.4	360.6±52.5	171.8±26.0	75.4±11.8
OB+curcumin (1.25 mg/kg)	502.5±28.5	210.4±14.5	378.9±69.4	173.8±20.4	73.5±11.3
OB+curcumin (2.5 mg/kg)	551.9±56.5	206.7±9.6	418.1±79.6	178.1±44.1	69.3±11.7
OB+curcumin (5 mg/kg)	595.1±38.8*	195.1±8.2	451.1±53.9	256.7±41.8	69.7±10.6
OB+curcumin (10 mg/kg)	615.3±36.7*	180.3±11.4	588.2±74.9*	263.9±35.8*	61.6±10.8
Sham+imipramine (10 mg/kg)	631.3±39.6**	174.8±11.6	668.5±113.0**	225.9±14.9	67.9±11.3

Data are expressed as mean±S.E.M. in units of ng/g (n=6–7).
# P<0.05, compared with sham+vehicle.
* P<0.05, compared with OB+vehicle.
** P<0.01, compared with OB+vehicle.

olfactory sensory receptors does not produce the characteristic symptoms of OB (Alberts and Friedman, 1972). Neuronal projections from the olfactory bulbs to the limbic system have a major influence on emotional behavior. Bilateral olfactory bulbectomy produces a well-characterized syndrome of behavioral and physiological changes that resemble clinical depression symptoms (Masini et al., 2004). Of the behavioral changes that occur following OB, two emerge that respond to antidepressant compounds in a selective fashion, namely the passive avoidance deficit and the open field hyperactivity (Harkin et al., 2003). Moreover, OB produces changes in the monoaminergic systems throughout the brain that resemble those seen in depression (Grecksch et al., 1997; Mudunkotuwa and Horton, 1996). Therefore, compared to other animal models, the OB rat appears to be a particularly applicable model for studying the neurobiological basis of depression and the mechanisms of action of antidepressant drugs.

Although most antidepressants alleviate immobility in the forced swim test after either acute or chronic treatment, the reversal of the behavioral deficits in the OB model requires chronic administration (Mudunkotuwa and Horton, 1996). Therefore, in the present study, we studied the effect of chronically administered curcumin. In addition, our pilot studies focused on the antidepressant effects of curcumin in naïve rodents (data not shown). Because antidepressant drugs do not show mood-elevating effects in normal healthy human subjects, it is necessary to assess the activity of this compound in animal models that simulate certain aspects of depression. The OB rat is one such model (Redmond et al., 1997). Chronic administration of curcumin did not result in any observable behavioral changes in the sham-operated groups in the open field and step-down passive avoidance tests, but attenuated the hyperactive response and passive avoidance deficit, in a dose-dependent manner, in the OB groups. Such interactions indicate that the behavioral effects of curcumin depend on the neuropharmacological state of an animal prior to administration. These curcumin effects were similar to imipramine, which was used in this study as a positive control. Moreover, the present results are also consistent with previous studies that tested the effects of antidepressant treatments in OB rats, such as desipramine, amitriptyline or mianserin (Iwasaki et al., 1986; Cairncross et al., 1979).

Antidepressant therapy includes various drugs with diverse pharmacological mechanisms as well as non-drug treatments (McLoughlin and Hodge, 1990). However neurobiological basic research, as well as clinical studies, have indicated that the monoamine systems (5-HT, NA and DA) are critically involved in the development of the clinical depression (Elhwuegi, 2004). Consistent with this view, most antidepressive drugs exert their action by elevating synaptic monoamine concentrations (Schloss and Henm, 2004). There are many studies indicating that OB reduces in 5-HT and NA and elevates 5-HIAA; these changes may mediate the behavioral abnormalities (e.g. hyperactivity and cognitive deficits) that resemble depression symptoms (Van Rijzingen et al., 1995). In the present experiment, we focused our interest on two distinct brain regions, the hippocampus and the frontal cortex. The involvement of these brain regions in emotional, motivational, and mnemonic processes may be related to their involvement in depression (Butterweck et al., 2002). It is interesting that Kelly et al. (1997) suggested that impaired hippocampal function and/or hippocampal edema may underlie memory deficits in OB animals. Magnetic resonance imaging has revealed alterations in signal intensity in the frontal cortex of OB rats and these alterations may contribute to the observed behavioral abnormalities (Wrynn et al., 2000). Hence, these two regions have been implicated in the behavioral alterations seen in depression, and changes in monoamine concentrations in these two regions may be relevant to clinical investigations of the depressed state.

The present results demonstrated that OB results in decreased 5-HT and NA levels together with increased 5-HIAA and DOPAC levels in the hippocampus. Similar changes were observed in the frontal cortex. Thus, the OB rat appears to be an appropriate model of hyposerotonergic and hyponoradrenergic depression (Lumia et al., 1992). Moreover, our data suggest that chronic curcumin administration (5 and 10 mg/kg) reverses OB-induced decreases in 5-HT and NA levels in both the hippocampus and the frontal cortex. Curcumin treatment also decreased 5-HIAA levels in the hippocampus and the frontal cortex in OB rats. These regional changes of monoamine levels as well as their metabolites (especially of 5-HT and NA) were similarly reversed by chronic treatment with imipramine (10 mg/kg).

Generally, the most widely accepted hypotheses of the biological basis of depression implicate NA or 5-HT system dysfunction. But the importance of the DA system in the pathophysiology and treatment of depression should not be ignored. Serra et al. (1979) proposed the involvement of dopaminergic presynaptic receptors in the action of antidepressants. In the present study, a significantly increased concentration of DA in the frontal cortex and a trend towards increased DA in the hippocampus were observed in OB rats that received 10 mg/kg curcumin. In addition, trends towards decreased DOPAC levels both in the hippocampus and the frontal cortex were observed in OB rats with 10 mg/kg curcumin.

Behavioral studies have suggested that reversing the 5-HT, NA and/or DA deficits in the brain are important for behavioral restoration in OB rats (Iwasaki et al., 1986). The present results strongly indicate that the behavioral changes following OB result from reductions of 5-HT, NA and/or DA, and further suggest that the effects of curcumin or imipramine on monoamine levels may be underlie the drugs' effects on behavior. Previous studies have indicated that curcumin inhibits the monoamine oxidase activity (Mazzio et al., 1998). Therefore, we believe from our findings that curcumin influenced, at least in part, the metabolism of the three monoamines, to restore normal monoaminergic function. Of course, other mechanisms may also increase the availability of brain monoamines, such as inhibition of monoamine re-uptake. Therefore, further experiments will be necessary to elucidate the mechanisms of the putative antidepressant effects of curcumin.

In conclusion, curcumin exerts antidepressant effects in both animal models studied here, and these effects may be mediated

by the central monoaminergic neurotransmitter systems. The convergence of these findings suggest that curcumin may be useful as a powerful, natural antidepressant agent.

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References

Alberts JR, Friedman MI. Olfactory bulb removal but not ansomia increases emotionality and mouse killing. Nature 1972;238:454–5.

Butterweck V, Bockers T, Korte B, Wittkowski W, Winterhoff H. Long-term effects of St John's wort and hypericin on monoamine levels in rat hypothalamus and hippocampus. Brain Res 2002;930:21–9.

Cairncross KD, Cox B, Foster C, Wren AT. Olfactory projection systems, drugs and behavior: a review. Psychoneuroendocrinology 1979;4:253–72.

Chen JX, Tang YT. Effect on xiaoyao powder on changes of relative brain zone CRF gene expression in chronic restrained stress rats. Chin J Appl Physiol 2004;20(1):71–4.

Dar A, Khatoon S. Behavioral and biochemical studies of dichloromethane fraction from the *Areca catechu* nut. Pharmacol Biochem Behav 2000;65(1):1–6.

Elhwuegi AS. Central monoamines and their role in major depression. Prog Neuro-Psychopharmacol 2004;28:435–51.

Franklin KBJ, Paxinos G. The mouse brain in stereotaxic coordinates. San Diego: Academic Press; 1997.

Grecksch G, Zhou D, Franke C, Schroder U, Sabel B, Becker A, et al. Influence of olfactory bulbectomy and subsequent imipramine treatment on 5-hydroxytryptaminergic presynapses in the rat frontal cortex: behavioral correlates. Br J Pharmacol 1997;122:1725–31.

Harkin A, Kelly JP, Leonard BE. A review of the relevance and validity of olfactory bulbectomy as a model of depression. Clin Neurosci Res 2003;3:253–62.

Iwasaki K, Fujiwara M, Shibata S, Ueki S. Changes in brain catecholamine levels following olfactory bulbectomy and the effect of acute and chronic administration of desipramine in rats. Pharmacol Biochem Behav 1986;24:1715–9.

Janscar S, Leonard BE. The olfactory bulbectomized rat as a model of depression. In: Usdin E, Goldstein M, Friedhoff AJ, Georgotas A, editors. Frontiers in neuropsychiatric research. New York: Macmillan Press; 1983. p. 357–72.

Kelly JP, Wrynn AS, Leonard BE. The olfactory bulbectomized rat as a model of depression: an update. Pharmacol Ther 1997;74(3):299–316.

Kong LD, Cheng CHK, Tan RX. Monoamine oxidase inhibitiors from rhizoma of *Coptis chinensis*. Planta Med 2001;67:74–6.

Lumia AR, Teicher MH, Slachli F, Ayers E, Possidente B. Olfactory bulbectomy as a model of agitated hyposerotonergic depression. Brain Res 1992;587:181–5.

Masini CV, Holmes PV, Freeman KG, Maki AC, Edwards GL. Dopamine overflow is increased in olfactory bulbectomized rats: an in vivo microdialysis study. Physiol Behav 2004;81(1):111–9.

Mazzio EA, Harris N, Soliman KF. Food constituents attenuate monoamine oxidase activity and peroxide levels in C6 astrocyte cells. Planta Med 1998;64(7):603–6.

McLoughlin IJ, Hodge JS. Zinc in depressive disorder. Acta Psychiatr Scand 1990;82(6):451–3.

Motterlini R, Foresti R, Bassi R, Green CJ. Curcumin, an antioxidant and anti-inflammatory agent, induces heme oxygenase-1 and protects endothelial cells against oxidative stress. Free Radic Biol Med 2000;28(8):1303–12.

Mudunkotuwa NT, Horton RW. Desipramine administration in the olfactory bulbectomized rat: changes in brain β-adrenoceptor and 5-HT_{2A} binding sites and their relationship to behavior. Br J Pharmacol 1996;117:1481–6.

Nitta A, Furukawa Y, Hayashi K, Hiramatsu M, Kameyama T, Hasegawa T, et al. Denervation of dopaminergic neurons with 6-hydroxydopamine increases nerve growth factor content in rat brain. Neurosci Lett 1992; 144(1–2):152–6.

Nowak G, Szewczyk B, Wieronska JM, Branski P, Palucha A, Pilc A, et al. Antidepressant-like effects of acute and chronic treatment with zinc in forced swimming test and olfactory bulbectomy model in rats. Brain Res Bull 2003;61:159–64.

Pilc A, Klodzinska A, Branski P, Nowak G, Palucha A, Szewczyk B, et al. Multiple MPEP administrations evoke anxiolytic-and antidepressant-like effects in rats. Neuropharmacology 2002;43:181–7.

Porsolt RD, Anton G, Blavet N, Jalfre M. Behavioral despair in rat: a new model sensitive to antidepressant treatment. Eur J Pharmacol 1978:379–91.

Redmond AM, Kelly JP, Leonard BE. Behavioral and neurochemical effects of dizocilpine in the olfactory bulbectomized rat model of depression. Pharmacol Biochem Behav 1997;58(2):355–9.

Redmond AM, Kelly JP, Leonard BE. The determination of the optimal dose of milnacipran in the olfactory bulbectomized rat model of depression. Pharmacol Biochem Behav 1999;62(4):619–23.

Schloss P, Henm FA. New insights into the mechanisms of antidepressant therapy. Pharmacol Ther 2004;102:47–60.

Serra G, Agriolas A, Klimek V, Fadda F, Gessa GL. Chronic treatment with antidepressants prevents the inhibitory effect of small doses of apomorphine on dopamine synthesis and motor activity. Life Sci 1979;25:415–23.

Thiyagarajan M, Sharma SS. Neuroprotective effect of curcumin in middle cerebral artery occlusion induced focal cerebral ischemia in rats. Life Sci 2004;74:969–85.

Van Rijzingen IM, Gispen WH, Spruijt BM. Olfactory bulbectomy temporarily impairs morris maze performance: an ACTH (4–9) analogue accelerates return of function. Physiol Behav 1995;58(1):147–52.

Vázquez-Palacios G, Bonilla-Jaime H, Velázquez-Moctezuma J. Antidepressant-like effects of the acute and chronic administration of nicotine in the rat forced swimming test and its interaction with flouxetine. Pharmacol Biochem Behav 2004;78:165–9.

Wrynn AS, Sweeney CPM, Franconi F, Lemaire L, Pouliquen D, Herlidou S, et al. An in-vivo magnetic resonance imaging study of the olfactory bulbectomized rat model of depression. Brain Res 2000;879:193–9.

Yu ZF, Kong LD, Chen Y. Antidepressant activity of aqueous extracts of curcuma longa in mice. J Ethnopharmacol 2002;83:161–5.