Toxicol. Res.
Vol. 26, No. 4, pp. 315-320 (2010)

Official Journal of
Korean Society of Toxicology

Available online at http://www.toxmut.or.kr

Toluene Induces Depression-Like Behaviors in Adult Mice

Miyoung Yang[1], Sung-Ho Kim[1], Jong-Choon Kim[1], Taekyun Shin[2] and Changjong Moon[1]

[1]*Departments of Veterinary Anatomy and Veterinary Toxicology, College of Veterinary Medicine, Chonnam National University, Gwangju 500-757*

[2]*Department of Veterinary Anatomy, College of Veterinary Medicine, Jeju National University, Jeju 690-756, Korea*

(Received October 12, 2010; Revised October 29, 2010; Accepted November 6, 2010)

It has been clinically reported that toluene causes mental depression in humans. However, the detrimental effects of toluene exposure on brain function and the relation between features of mental depression and toluene exposure are poorly understood. This study evaluated depression-like behaviors in adult C57BL/6 mice after administration of toluene, and elucidated the effects of classical antidepressants on the depression-like behaviors. For the estimation of depression-like behaviors, tail suspension test (TST) and forced-swim test (FST) were performed 1, 4 and 16 days after toluene (0~1000 mg/kg bw) treatment. In addition, classical antidepressants such as fluoxetine (FLX, 20 mg/kg bw) and imipramine (IMI, 40 mg/kg bw) were administered 12 h and 1 h before the tests. In the TST and FST, toluene-treated mice exhibited a longer duration of immobility than vehicle-treated mice 1 and 4 days after toluene treatment. The depression-like behaviors were significantly reversed by FLX and IMI. The weight of the adrenal gland and the size of adrenocortical cells were significantly higher in toluene-treated mice compared to vehicle-treated controls. It is suggested that acute toluene exposure of adult mice is sufficiently detrimental to induce depression. In addition, this study has established a mouse model for a depressive state induced by toluene treatment.

***Key words*:** Toluene, Depression, Behavior, Adrenal gland, Animal model

INTRODUCTION

Depression is a serious and incapacitating disorder with a heavy social burden that carries a substantial lifetime risk (Greenberg *et al.*, 2003; Millan, 2004). Severe forms of depression affect 2~5% of the U.S. population, and mood disorders affect 7% of the world's population and rank among the top 10 causes of disability (Murray and Lopez, 1996). Work-related injuries contribute to the development of psychopathologies such as depression (Stice and Dik, 2009). Work-related depression has emerged as a major cause of long-term sickness. The relationship between work and depression is bidirectional: work gives acceptance and self-confidence to the individual, but workplace stress may precipitate depression (Unger, 2007). Despite its status as the most common psychiatric disorder, depression is poorly understood in terms of the mechanisms.

Toluene is a volatile organic compound that is widely used as a paint-thinner, industrial degreasing agent, and dry-cleaning agent. However, toluene is associated with neurophysiological and psychological disturbances (Grasso *et al.*, 1984), and it is considered an important health hazard (Fishbein, 1985). Chronic toluene intoxication in humans leads to development of symptoms such as palpitation, insomnia, dizziness with headache, memory impairment, euphoria while working, and mental depression during the weekend relief from work-related toluene exposure (Lee *et al.*, 2003). Similar to those of other sedative-hypnotics, toluene can readily cross the blood-brain barrier and produce central nervous system effects (Balster, 1998). In animal experiments, toluene exposure leads to changes in neurobehavioral and neurobiological functions (Berenguer *et al.*, 2003; Kondo *et al.*, 1995; Reigel and French, 1999; Seo *et al.*, 2010; von Euler *et al.*, 2000). However, little is known about the precise mechanisms of depression in humans and in experimental animal models after toluene exposure.

This study focused on depression-like behaviors using the tail suspension test (TST) and forced swim test (FST) in adult C57BL/6 mice after the administration of toluene. To examine the relationships between toluene-induced depression-like behaviors and stress, the changes of several stress-

Correspondence to: Changjong Moon, Department of Veterinary Anatomy, College of Veterinary Medicine, Chonnam National University, 300 Yongbong-dong, Buk-gu, Gwangju 500-757, Korea
E-mail: moonc@chonnam.ac.kr

related parameters were examined after toluene treatment. Additionally, the effects of antidepressants on toluene-induced depression-like behaviors were evaluated.

MATERIALS AND METHODS

Animals. Male C57BL/6 mice aged 8~9 weeks (Orient Bio, Gyunggi-do, Korea) were housed in a room that was maintained at 23 ± 2°C, relative humidity of 50 ± 5%, artificial lighting from 08:00~20:00 h and with 13~18 air changes hourly. The animals were given tap water and commercial rodent chow (Samyang Feed, Seoul, Korea) *ad libitum*. All animal experiments followed a protocol approved by the Committee for Animal Experimentation at Chonnam National University, and the animals were cared for in accordance with the Guidelines for Animal Experiments.

Toluene administration and tissue sampling. Toluene (HPLC grade, 99.8% pure) was obtained from Junsei Chemical (Tokyo, Japan). Toluene-treated mice received an intraperitoneal (i.p.) injection of toluene dissolved in corn oil (500 mg/kg bw). Vehicle-treated control mice were injected with only corn oil. I.p. injection of toluene produces the same behavioral symptoms as inhalation (Kondo *et al.*, 1995; Reigel and Frech, 1999) and is easy to handle. In addition, the dosages were selected on the basis of a previous study, which demonstrated the no dosage-related body weight loss and effect on basic locomotor activity (Seo *et al.*, 2010).

The behavior tests were performed at 1, 4 and 16 days after vehicle or toluene administration. To observe the dose-dependent effects of toluene on locomotor activity and depression-like behaviors, mice were administered 0 (vehicle-treated control), 100, 500, or 1000 mg/kg bw dose of toluene, and the behavior tests were performed at 4 days after treatment. In addition, to examine the effect of antidepressants on toluene-induced depression-like behaviors, classical antidepressants, such as imipramine hydrochloride (IMI, 40 mg/kg bw; Sigma-Aldrich, St. Louis, MO, USA) and fluoxetine hydrochloride (FLX, 20 mg/kg bw; Sigma-Aldrich) were dissolved in saline and intraperitoneally administrated 12 h and 1 h, respectively, prior to the behavioral tests.

Body weights of mice were measured 4 days after vehicle and toluene injection, and then the animals were euthanized by ether inhalation. Blood was collected in heparinized tubes and plasma was separated. At dissection, the adrenal gland was also weighed.

For measurement of the cell size of adrenocortical cells, both adrenal glands from each mouse were fixed with 10% neutral-buffered formalin. The tissue sections were stained with hematoxylin and eosin (H&E).

Open field test. Open-field analysis was used to measure the activity of the mice in a novel environment. Parameters including ambulatory movement episodes, total moving distance (cm), ambulatory movement time (s), and rest time (s) were determined over 5 min using the TruScan Photo Beam Activity System (Coulbourn Instruments, Whitehall, PA, USA).

TST. The TST was similar to that described by Steru *et al.* (1985). Briefly, mice were suspended from a plastic rod mounted 50 cm above the surface by fastening the tail to the rod with adhesive tape. Immobility was measured for 6 min. Immobility was defined as the absence of any limb or body movements, except those caused by respiration.

FST. The FST was similar to that described by Porsolt *et al.* (1977). Briefly, mice were gently placed in a clear plastic cylinder with a diameter of 13 cm and a height of 23 cm that was filled with 10 cm of clear water at 23~25°C. The test duration was 6 min, and immobility was measured during the last 4 min. Immobility was defined as the absence of any horizontal or vertical movement in the water, but excluded minor movements required for the mouse to keep its head above the surface. The water was replaced before each animal began the test.

Measurement of the size of adrenocortical cells. Images of adrenal gland sections were taken with a digital camera mounted on a microscope (Leica DM IRBE; Leica Micro systems GmbH, Wetzlar, Germany). The size of adrenocortical cells in the zona fascicularis was determined using Leica QWin image analyzing software (Leica Microsystems). Two fields (× 100) in each sample were measured for the vehicle- and toluene-treated groups.

Quantitative analysis of serum corticosterone. The measurement of serum corticosterone was performed 4 days after administration of vehicle or toluene. Blood was collected from vehicle- and toluene-treated mice, and was separated into blood cells and serum. The concentration of serum corticosterone was measured according to the manufacturer's instructions using a Corticosterone Enzyme Immunoassay kit (Assay Designs, Ann Arbor, MI, USA).

Statistical analysis. The data are reported as the mean ± SE and were analyzed using a one-way analysis of variance (ANOVA) followed by a Student-Newman-Keuls post hoc test for multiple comparisons. In all cases, a p value < 0.05 was considered significant.

RESULTS

Time-related effect of toluene on depression-like behaviors. TST and FST have been recognized as useful experimental paradigms for assessing depression-like behavior and the activity of antidepressants. These tests were performed at 1, 4 and 16 days after injection of 500 mg/kg bw

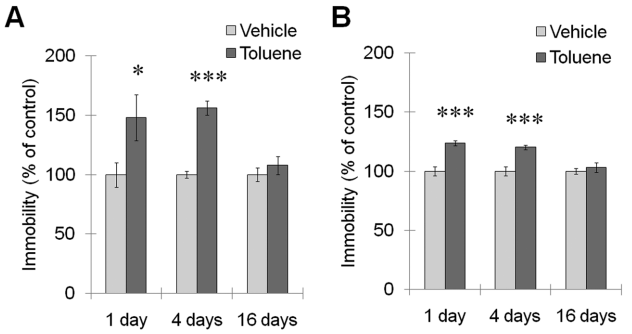

Fig. 1. Time-dependent effect of toluene injection on immobility measured during the TST (A) and FST (B) in adult mice. Vehicle (corn oil) or toluene (500 mg/kg bw) was injected at 1 day, 4 days and 16 days before the test. (A) In the TST, the immobility (% of control) of mice 1 day and 4 days after toluene injection was significantly higher in toluene-treated mice than in vehicle-treated control mice. However, mice 16 days after toluene injection showed no significant difference in immobility. (B) In the FST, the immobility (% of control) in mice was significantly higher in mice 1 day and 4 days after toluene injection than in vehicle-treated control mice, but not in mice 16 days after toluene injection, similar to the results of TST. The values reported are the mean ± SE ($n = 7$ per group). * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ *vs.* vehicle-treated controls.

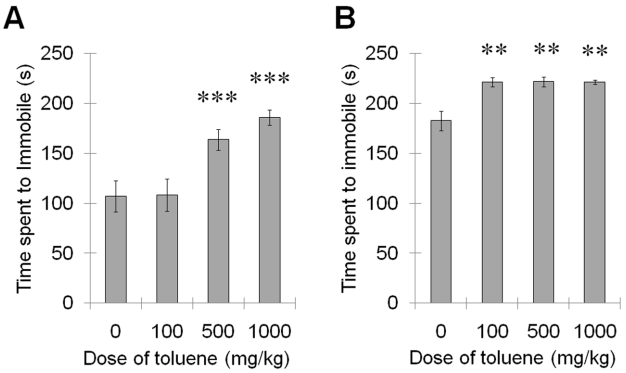

Fig. 2. Dose-dependent effect of toluene injection on immobility measured during the TST (A) and FST (B) in adult mice. Vehicle (corn oil) or toluene (100, 500 and 1000 mg/kg bw) was injected 4 days before the test. (A) In the TST, mice injected 100 mg/kg bw toluene showed similar time spent to immobile (s) with vehicle controls. However, mice injected 500 and 1000 mg/kg bw toluene significant showed higher time spent to immobile than vehicle-treated control mice. (B) In the FST, mice injected toluene in all of selected doses showed higher time to spent immobile than vehicle-treated control mice. The values (seconds) reported are the mean ± SE ($n = 5$ per group). ** $p < 0.01$, *** $p < 0.001$ *vs.* vehicle-treated controls.

toluene to examine time-related depression-like behaviors of toluene. Toluene-treated mice exhibited a longer duration of immobility during the TST compared to vehicle-treated mice at post-injection day 1 (vehicle-treated control: 92.7 ± 9.5 sec, toluene-treated mice: 137.3 ± 17.9 sec; $n = 7$, $p < 0.05$) and 4 (vehicle-treated control: 120.7 ± 3.4 sec, toluene-treated mice: 188.6 ± 7.1 sec; $n = 7$, $p < 0.001$) (Fig. 1A). However, 16 days after toluene injection, no significant difference was found in immobility time of the TST compared to vehicle controls (vehicle-treated control: 181.9 ± 10.2 sec, toluene-treated mice: 195.9 ± 13.7 sec; $n = 7$, $p = 0.429$; Fig. 1A). Similarly, the immobility time in the FST increased significantly in toluene-treated mice as compared to the vehicle-treated control group at post-injection day 1 (vehicle-treated control: 167 ± 6.5 sec, toluene-treated mice: 206.6 ± 3.3 sec; $n = 7$, $p < 0.001$) and 4 (vehicle-treated control: 181.3 ± 7.1 sec, toluene-treated mice: 218 ± 3.4 sec; $n = 7$, $p < 0.001$), whereas mice examined 16 days after toluene injection did not show depression-like behavior (vehicle-treated control: 191.3 ± 4.9 sec, toluene-treated mice: 197.6 ± 7.7 sec; $n = 7$, $p = 0.503$) (Fig. 1B).

Dose-related effect of toluene on depression-like behaviors. TST and FST were performed at 4 days after toluene injection (0~1000 mg/kg bw) to examine dose-related depression-like behaviors. As shown in Fig. 2A, the TST-determined immobility time increased progressively with increasing dose of injected toluene (0~1000 mg/kg bw), although there was no significant difference up to 100 mg/kg bw toluene. In FST, the immobility time significantly increased at 100, 500 or 1000 mg/kg bw of toluene compared to vehicle controls (0 mg/kg bw), but leveled off slowly as the dose was increased (Fig. 2B).

Effect of toluene in the locomotion in the open-field test. Basic locomotor activity of mice was first examined 4 days after toluene treatment (0~1000 mg/kg bw) in a novel open-field analysis. The analysis quantified overall locomotor activity that could affect motivation and performance of the mice. Vehicle-treated (control) and toluene-treated mice showed comparable moving distance, ambulatory movement time, ambulatory movement count and resting time (Table 1). This indicates that toluene exposure does not alter locomotor activity significantly in adult mice.

Effect of antidepressants on toluene-induced depression-like behaviors. Toluene significantly increased immobility time in TST and FST. Further analyses examined the effects of classical antidepressants, IMI and FLX to confirm toluene-induced increases of immobility in TST- and FST-related depression. Each test was performed at 4 days after injection of vehicle or toluene (500 mg/kg bw), and antidepressants were injected (i.p.) 12 h and 1 h before testing. Both IMI and FLX significantly decreased the toluene-induced increased immobility time in TST (IMI: 59.8 ± 16.7 sec, $p < 0.0001$; FLX: 43.7 ± 11.0 sec, $p < 0.0001$ *vs.* toluene-treated group: 197.0 ± 6.9 sec, $n = 6$) (Fig. 3A). In FST, IMI and FLX significantly reversed the toluene-induced

Table 1. Open-field analysis of mice placed in a novel environment at 4 days after vehicle (0 mg/kg bw) and toluene (100~1000 mg/kg bw) injection

	Toluene dose (mg/kg bw)			
	0	100	500	1000
Movement episodes	29.2 ± 2.67	27.0 ± 2.85	25.4 ± 2.68	33.6 ± 5.26
Distance (cm)	448.1 ± 88.6	466.8 ± 10.6	470.6 ± 28.0	387.2 ± 35.1
Movement time (sec)	225.8 ± 5.66	233.4 ± 3.31	236.2 ± 3.40	211.0 ± 9.29
Rest time (sec)	38.8 ± 2.48	36.0 ± 4.49	30.4 ± 3.87	44.4 ± 6.64

The data are reported as the mean ± SE ($n = 5$ per group).

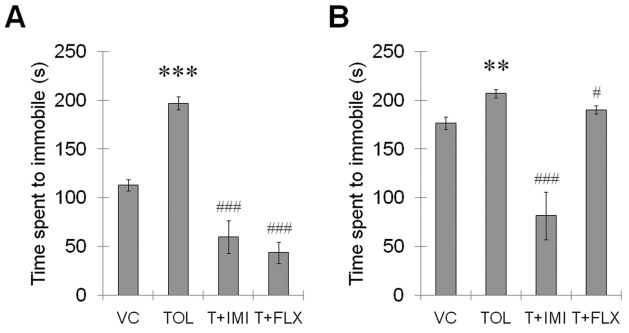

Fig. 3. The effect of antidepressant (IMI and FLX) on depression-like behaviors induced toluene (500 mg/kg bw) injection in TST (A) and FST (B). (A) In TST, IMI and FLX significantly reduced immobile time (seconds) that increased by toluene injection. Furthermore, IMI- and FLX-treated mice before toluene injection showed lower time to spent to immobile than vehicle-treated control mice. (B) In addition, IMI and FLX significantly declined immobile time that increased by toluene injection in FST. However, the effect of IMI was stronger than of FLX to be prolonged immobile time induced toluene. IMI and FLX reversed depression-like behaviors induced toluene injection. ** $p < 0.01$, *** $p < 0.001$ *vs.* vehicle-treated controls. # $p < 0.05$, ### $p < 0.001$ *vs.* toluene-treated controls.

depression-like behavior. However, the effects of the two antidepressants differed, in that IMI strongly decreased the immobility time, whereas FLX produced only a marginally significant decrease (IMI: 81.5 ± 24.7 sec, $p < 0.001$; FLX: 190.2 ± 4.29 sec, $p < 0.05$ *vs.* toluene-treated group: 207.0 ± 4.33 sec, $n = 6$) in contrast to the effects in TST (Fig. 3B).

Effect of toluene on stress-related parameters. Total adrenal gland weight per body weight, cortical cell size and concentration of corticosterone were examined as stress-related parameters to evaluate the effect of toluene (0~1000 mg/kg bw) injection (Table 2). All the doses of toluene significantly induced increased adrenal weight per body weight (μg/mg) (100 mg/kg bw: 242.3 ± 8.88, $p < 0.05$; 500 mg/kg bw: 234.7 ± 8.86, $p < 0.05$; 1000 mg/kg bw: 256.0 ± 9.39, $p < 0.01$ *vs.* vehicle-treated control: 206.6 ± 8.43, $n = 9$) 4 days after injection compared to vehicle-treated controls. Similarly, cortical cell size was also significantly increased by all selected doses (100 mg/kg bw: $52.1 \pm 2.27 \times 10^2$ μm^2, $p < 0.01$; 500 mg/kg bw: $50.0 \pm 3.02 \times 10^2$ μm^2, $p < 0.05$; 1000 mg/kg bw: $57.0 \pm 2.19 \times 10^2$ μm^2, $p < 0.0001$ *vs.* vehicle-treated control, $n = 9$) 4 days after injection compared with vehicle-treated controls ($40.9 \pm 1.72 \times 10^2$ μm^2, $n = 9$). In the toluene-treated mice, H&E staining revealed that cells were significantly enlarged, especially the cytoplasm, compared to vehicle-treated controls (Fig. 4). Furthermore, the changes in concentration of serum corticosterone were more pronounced in cells treated with all doses of toluene, as compared to vehicle-treated control mice 4 days after injection, although the changes were not statistically significant (Table 2).

DISCUSSION

Presently, toluene exposure induced depression-like behaviors as measured by TST and FST in adult mice. In addition, these depression-like behaviors induced toluene exposure reverse by two classical antidepressants. This suggests that toluene exposure causes depression in adult mice.

Long-term recreational, occupational, as well as environmental exposure to toluene may result in a variety of neurological manifestations, including ataxia, dementia, sensory

Table 2. Changes of the examined markers between vehicle-treated control group (0 mg/kg bw) and toluene-treated group (100~1000 mg/kg bw) at 4 days after injection (% of control)

	Toluene dose (mg/kg bw)			
	0	100	500	1000
Adrenal weight per body weight	100 ± 4.08	117.2 ± 4.30*	113.6 ± 4.29*	123.9 ± 4.54**
Cortical cell size	100 ± 4.19	123.7 ± 4.92**	116.3 ± 5.08*	135.5 ± 4.36***
Corticosterone	100 ± 31.60	146.4 ± 24.39	139.1 ± 21.14	143.1 ± 33.30

The data are reported as the mean ± SE ($n = 9$ per group). * $p < 0.05$, ** $p < 0.01$ *** $p < 0.001$ *vs.* vehicle-treated controls.

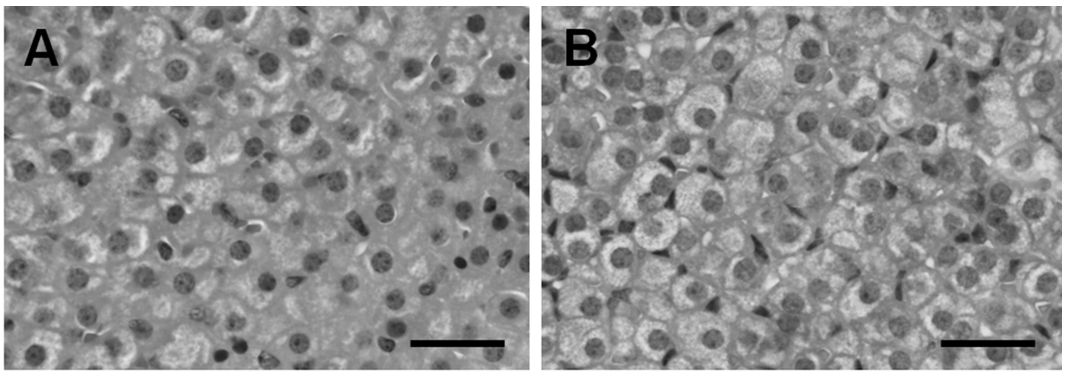

Fig. 4. Histological results by hematoxylin and eosin (H&E) staining for the vehicle-treated controls 4 days after injection with only corn oil (A) and toluene-treated mouse adrenal gland at 4 days administration of 1000 mg/kg bw toluene (B). In the toluene-treated group (B), the expansion of the adrenal cortical cells appeared compared with vehicle-treated controls (A). $n = 9$ per group. Scale bars in A and B represent 30 μm.

dysfunction, seizure, tremor and cognitive impairment, as well as depression (Benignus, 1981; Anderson and Loomis, 2003). Recently, Crez *et al.* (2009) reported on the antidepressant-like actions of toluene 30 min after inhalation. In contrast, another study reported that hippocampal dysfunctions such as cognitive impairment and depression were evident up to 4 days following toluene exposure (Seo *et al.*, 2010). The present data confirmed that toluene exposure induces depression-like behaviors in the short-term (1 and 4 days) reversibly, but not in the long-term (16 days) after acute toluene exposure in mice. This may reflect a time-dependent effect of toluene exposure.

Depression is a highly ubiquitous, complex and heterogeneous disorder with serious physical, mental and socioeconomical consequences. Among the mechanisms associated with depression, the role of stress in psychiatric disorders has been well-demonstrated; in particular, epidemiological data have lent strong support to the idea that stressful life events play a role in the etiology of depression (Kendler *et al.*, 1995). However, the precise mechanisms of the depression-like behaviors induced by toluene remain unknown. Here, two possible mechanisms can be offered. One theory posits that the genesis of depression is reduced brain plasticity. According to this theory, depression may be due not only to the changes in neurotransmitter concentrations and receptor activity levels, but also to impaired brain plasticity and tissue remodeling, and alterations in adult hippocampal neurogenesis (Kim *et al.*, 2008). The hippocampus is one of several limbic structures that have been extensively studied in individuals with learning and memory difficulties, and depression (Kim *et al.*, 2008; Seo *et al.*, 2010; Yang *et al.*, 2010). Toluene exposure in adult mice may reduce hippocampal neurogenesis and causes hippocampal dysfunctions such as depression and cognitive impairment (Seo *et al.*, 2010). Therefore, this reduction of neurogenesis by toluene exposure alters brain plasticity, and may induce hippocampal dysfunction, including depression.

Another mechanism is stress-related depression. Toluene exposure induces adrenocortical hypertrophy via the stress-responsive hypothalamus-pituitary-adrenal gland (HPA) axis, neither stimulating nor damaging adrenal cells directly (Gotohda *et al.*, 2005). As previously reported, the present study observed the significant enlargement of the adrenal gland, especially adrenocortical cells, upon toluene exposure. Additionally, the concentration of corticosterone was elevated, albeit non-significantly. Also, toluene induces the impairment of energy metabolism, oxidative stress in the prefrontal cortex and hippocampus-related chronic stress (Tagliari *et al.*, 2010). Therefore, toluene may induce depression via direct facilitation of the corticosterone excretion, or indirect damage to hippocampus such as impaired energy metabolism and oxidative stress. However, further studies should clarify the molecular and cellular mechanisms for the *in vivo* depression effects of toluene exposure.

In conclusion, toluene exposure transiently induces depression-like behaviors in TST and FST, and these behavioral effects are reversed by antidepressants, suggesting that toluene exposure induces depression in adult C57BL/6 mice. Additionally, the behavioral effects are related with several stress-related parameters, such as weight of adrenal glands, size of adrenocortical cells and concentration of corticosterone. This study has established a mouse model of a depressive state induced by toluene treatment. This suggests that acute toluene treatment in mice is a beneficial animal model for screening new antidepressants and clarifying the precise mechanisms of mental depression.

ACKNOWLEDGMENTS

This work was supported by the Grant of the Korean Ministry of Education, Science and Technology (The Regional Core Research Program/Biohousing Research Institute). This

work was supported by the Biohousing Research Center.

REFERENCES

Anderson, C.E. and Loomis, G.A. (2003). Recognition and prevention of inhalant abuse. *Am. Fam. Physician*, **68**, 869-874.

Aonurm-Helm, A., Jurgenson, M., Zharkovsky, T., Sonn, K., Berezin, V., Bock, E. and Zharkovsky, A. (2008). Depression-like behaviour in neural cell adhesion molecule (NCAM)-deficient mice and its reversal by an NCAM-derived peptide, FGL. *Eur. J. Neurosci.*, **28**, 1618-1628.

Balster, R.L. (1998). Neural basis of inhalant abuse. *Drug Alcohol Depend.*, **51**, 207-214.

Benignus, V.A. (1981). Health effects of toluene: a review. *Neurotoxicology*, **2**, 567-588.

Berenguer, P., Soulage, C., Perrin, D., Pequignot, J.M. and Abraini, J.H. (2003). Behavioral and neurochemical effects induced by subchronic exposure to 40 ppm toluene in rats. *Pharmacol. Biochem. Behav.*, **74**, 997-1003.

Cruz, S.L., Soberanes-Chávez, P., Páez-Martinez, N. and López-Rubalcava, C. (2009). Toluene has antidepressant-like actions in two animal models used for the screening of antidepressant drugs. *Pychopharmacology (Berl)*, **204**, 279-286.

Fishbein, L. (1985). An overview of environmental and toxicological aspets of aromatic cydrocarbons II. *Sci. Total Environ.*, **42**, 267-288.

Grasso, P., Sharratt, M., Davies, D.M. and Irvine, D. (1984). Neurological and psychological disorders and occupational exposure to organic solvents. *Food Chem. Toxicol.*, **22**, 819-852.

Greenberg, P.E., Leong, S.A., Birnbaum, H.G. and Robinson, R.L. (2003). The economic burden of depression with painful symptoms. *J. Clin. Psychiatry*, **64**, 17-23.

Gotohda, T., Tokunaga, I. and Kubo, S. (2005). Toluene inhalation-induced adrenocortical hypertrophy and endocrinological changes in rat. *Life Sci.*, **76**, 1929-1937.

Hsieh, G.C., Sharma, R.P. and Parker, R.D.R. (1991). Hypothalamic-pituitary-adrenocortical axis activity and immune function after oral exposure to benzene and toluene. *Immunopharmacology*, **21**, 23-32.

Kendler, K.S., Kessler, R.C., Walters, E.E., MacLean, C., Neale, M.C., Heath, A.C. and Eaves, L.J. (1995). Stressful life events, genetic liability, and onset of an episode of major depression in women. *Am. J. Psychiatry*, **152**, 833-842.

Kim, J.S., Lee, H.J., Kim, J.C., Kang, S.S., Bae, C.S., Shin, T., Jin, J.K., Kim, S.H., Wang, H. and Moon, C. (2008). Transient impairment of hippocampus-dependent learning and memory in relatively low-dose of acute radiation syndrome is associated with inhibition of hippocampal neurogenesis. *J. Radiat. Res.*, **49**, 517-526.

Kondo, H., Huang, J., Ichihara, G., Kamijima, M., Saito, I., Shibata, E., Ono, Y., Hisanaga, N., Takeuchi, Y. and Nakahara, D. (1995). Toluene induces behavioral activation without affecting striatal dopamine metabolism in the rat: behavioral and microdialysis studies. *Pharmacol. Biochem. Behav.*, **51**, 97-101.

Lee, Y.L., Pai, M.C., Chen, J.H. and Guo, Y.L. (2003). Central neurological abnormalities and multiple chemical sensitivity caused by chronic toluene exposure. *Occup. Med. (Lond)*, **53**, 479-482.

Millan, M.J. (2004). The role of monoamines in the actions of established and "novel" antidepressant agents: a critical review. *Eur. J. Pharmacol.*, **500**, 371-384.

Murray, C.J. and Lopez, A.D. (1996). Evidence-based health policy - lessons from the Global Burden of Disease Study. *Science*, **274**, 740-743.

Porsolt, R.D., Bertin, A. and Jalfre, M. (1977). Behavioural despair in mice: a primary screening test for antidepressants. *Arch. Int. Pharmacodyn. Ther.*, **229**, 327-336.

Riegel, A.C. and French, E.D. (1999). An electrophysiological analysis of rat ventral tegmental dopamine neuronal activity during acute toluene exposure. *Pharmacol. Toxicol.*, **85**, 37-43.

Seo, H.S., Yang, M., Song, M.S., Kim, J.S., Kim, S.H., Kim, J.C., Kim, H., Shin, T., Wang, H. and Moon, C. (2010). Toluene inhibits hippocampal neurogenesis in adult mice. *Pharmacol. Biochem. Behav.*, **94**, 588-594.

Steru, L., Chermat, R., Thierry, B. and Simon, P. (1985). The tail suspension test: a new method for screening antidepressants in mice. *Psychopharmacology (Berlin)*, **85**, 367-370.

Stice, B.D. and Dik, B.J. (2009). Depression among injured workers receiving vocational rehabilitation: contributions of work values, pain, and stress. *J. Occup. Rehabil.*, **19**, 354-363.

Tagliari, B., Noschang, C.G., Ferreira, A.G.K., Ferrari, O.A., Feksa, L.R., Wannmacher, C.M.D., Dalmaz, C. and Wyse, A.T.S. (2010). Chronic variable stress impairs energy metabolism in prefrontal cortex and hippocampus of rats: prevention by chronic antioxidant treatment. *Metab. Brain Dis.*, **25**, 169-176.

Unger, H.P. (2007). Work-related depression. *Psychiatr. Prax.*, **34**, S256-260.

von Euler, M., Pham, T.M., Hillefors, M., Bjelke, B., Henriksson, B. and von Euler, G. (2000). Inhalation of low concentrations of toluene induces persistent effects on a learning retention task, beam-walk performance, and cerebrocortical size in the rat. *Exp. Neurol.*, **163**, 1-8.

Yang, M., Kim, J.S., Song, M., Kim, S.H., Kang, S.S., Bae, C.S., Kim, J.C., Wang, H., Shin, T. and Moon, C. (2010). Cyclophosphamide impairs hippocampus dependent learning and memory in adult mice: Possible involvement of hippocampal neurogenesis in chemotherapy-induced memory deficits. *Neurobiol. Learn. Mem.*, **93**, 487-494.