Psychiatry Research 271 (2019) 328–334

Contents lists available at ScienceDirect

Psychiatry Research

journal homepage: www.elsevier.com/locate/psychres

BDNF mediates the protective effects of scopolamine in reserpine-induced depression-like behaviors via up-regulation of 5-HTT and TPH1

Hanjie Yu[a,b,c,d,1], Dan Lv[a,b,c,d,1], Mengxin Shen[a,b,c,d,1], Yanhua Zhang[a,b,c,d,1], Dongsheng Zhou[a,b,*], Zhongming Chen[a,b,*], Chuang Wang[a,b,c,d,*]

[a] Ningbo Kangning Hospital, Ningbo, Zhejiang 315201, China
[b] Ningbo Key Laboratory of Behavioral Neuroscience, Ningbo University School of Medicine, 818 Fenghua Road, Ningbo, Zhejiang 315211, China
[c] Zhejiang Provincial Key Laboratory of Pathophysiology, Ningbo University School of Medicine, 818 Fenghua Road, Ningbo, Zhejiang 315211, China
[d] Department of Physiology and Pharmacology, Ningbo University School of Medicine, 818 Fenghua Road, Ningbo, Zhejiang 315211, China

ARTICLE INFO

Keywords:
Scopolamine
Depression
Serotonin transporter (5-HTT)
Brain-derived neurotrophic factor (BDNF)
Tryptophan hydroxylase 1 (TPH1)

ABSTRACT

Reserpine treatment in rodents has been shown to induce depression-like behaviors that mimic monoamine dysfunction implicated in the development of depression. Herein, we aimed to demonstrate the antidepressant-like activities of scopolamine, the muscarinic receptor antagonist, in a reserpine-induced mouse model. Mice were injected with 1.5 mg/kg (i.p.) of reserpine for 10 days, and the depression-like state was confirmed via the open field test (OFT) and forced swimming test (FST). Then, the mice were treated with scopolamine (25 μg/kg, i.p.) or saline for 3 days. Ten days of reserpine treatment resulted in a significant decrease in locomotor activity and an increase in immobility time in the OFT and FST, respectively, indicating that ten days of reserpine administration significantly induced depression-like behaviors in mice. However, scopolamine rapidly ameliorated the increase in immobility time in the FST and had no effect on locomotor activity in the OFT. In addition, the reserpine-induced decreases in serotonin transporter (5-HTT), brain-derived neurotrophic factor (BDNF) and tryptophan hydroxylase 1 (TPH1) in mouse hippocampus and prefrontal cortex (PFC) were significantly reversed by scopolamine. Our study provides evidence that scopolamine rapidly attenuates reserpine-induced depression in mice partially by regulating 5-HTT, BDNF and TPH1 in the hippocampus and PFC of mice.

1. Introduction

Depression is a common psychiatric illness that has been established to be associated with a decrease in serotonin and/or serotonin transporters in the brain (Pérez-Olmos et al., 2016; Bregman et al., 2017; Li et al., 2017). Notably, the pathophysiology of depression is strongly linked to impairments in serotonin (5-HT) neurotransmission (Khnychenko et al., 2017; Sivolap, 2017). The 5-HT released from serotonergic terminals is selectively taken up from the synaptic cleft into these terminals via the serotonin transporter (5-HTT) (Hansson et al., 1998), indicating that 5-HTT may serve as a physiologic mediator in depression. Additionally, growing evidence that 5-HTT has a strong association with depression in clinical (Daniele et al., 2011; Newberg et al., 2012; Dannlowski et al., 2014) and preclinical studies (Tang et al., 2013, 2014). The association between 5-HTT and depression warrants further investigation to elucidate mechanisms and identify potential therapeutic target.

It has been reported that reserpine irreversibly inhibit the vesicular uptake of monoamines, and recent study (Ikram and Haleem, 2017) suggest that repeated administration of reserpine could be serve as a progressive model of depression and could be used as a convenient and economic animal model for the face validity of antidepressant compounds. Importantly, a previous study (Xiao et al., 1999) found that chronic treatment with reserpine decreased 5-HTT mRNA levels in the rodent brain, supporting the use of reserpine treatment in rodents as a progressive animal model of depression. Recently, brain-derived neurotrophic factor (BDNF) has been shown to promote the survival and morphological differentiation of 5-HT neurons both in culture and in vivo (Eaton and Whittemore, 1996; Rumajogee et al., 2002). Additionally, substantial evidence suggests that BDNF also promotes the expression of tryptophan hydroxylase (TPH1) (the rate-limiting enzyme in 5-HT synthesis) (Siuciak et al., 1998; Goggi et al., 2002). In addition,

* Corresponding authors at: Ningbo Key Laboratory of Behavioral Neuroscience, Ningbo University School of Medicine, 818 Fenghua Road, Ningbo, Zhejiang 315211, China.
E-mail addresses: wyzhouds@sina.com (D. Zhou), chenzhongming@hotmail.com (Z. Chen), wangchuang@nbu.edu.cn (C. Wang).
[1] These authors contributed equally to this work..

https://doi.org/10.1016/j.psychres.2018.12.015
Received 30 August 2018; Received in revised form 18 November 2018; Accepted 3 December 2018
Available online 03 December 2018
0165-1781/

data from experiments using cell culture demonstrated a dose-dependent decrease in 5-HT uptake following exposure to BDNF (Mössner et al., 2000), in spite of a marked increase in mRNA levels of the 5-HTT (Rumajogee et al., 2002). Importantly, it was shown that BDNF-deficient mice have a reduced ability to clear 5-HT (Daws et al., 2007), indicating that the 5-HTT was regulated by BDNF.

Mechanistic studies should be conducted to demonstrate whether the reserpine-induced depression-like behaviors and changes of 5-HTT and TPH1 were alleviated by antidepressants produced BDNF up-regulation.

Interestingly, clinical trial (Park et al., 2018) and animal experiments (Ghosal et al., 2018) have been revealed that subclinical dose of scopolamine produces a rapid antidepressant-like effects. Importantly, the antidepressant-like actions of scopolamine in rodent models may depend on the level of BDNF in the brain (Rigal et al., 2016; Dong et al., 2017; Yu et al., 2017; Ghosal et al., 2018). However, whether the decrease of 5-HTT induced by reserpine could be reversed by scopolamine remains unknown. Our aim in the current study was to demonstrate whether scopolamine could rescue the reserpine-induced depression-like behaviors in mice by changing the levels of BDNF and then regulating TPH1 and 5-HTT levels in the mouse hippocampus and prefrontal cortex (PFC).

2. Methods

2.1. Animals

Adult male ICR mice (20–22 g) were born and reared in the animal facility of Ningbo University Medical School, China. All animals were maintained at 23 ± 2 °C and 60% ± 5% relative humidity under a 12-hour light/12-hour dark cycle (lights on at 07:00 AM) with ad libitum access to food and water. All procedures involving animals were conducted following the National Institute of Health (NIH) Guidelines for the Care and Use of Laboratory Animals (NIH Publications No. 80–23, revised 1996) as well as the European Community Council Directive for the Care and Use of Laboratory Animals of September 22, 2010 (2010/63/EU). All of the experiments were approved by the Institutional Animal Care and Use Committee of the Medical School of Ningbo University.

2.2. Drugs

Scopolamine (25 μg/kg, i.p.; Sigma-Aldrich, St. Louis, MO, USA) and reserpine (1.5 mg/kg, i.p.; Sigma-Aldrich, St. Louis, MO, USA) were dissolved in saline. Fig. 1A shows a schematic representation of the treatment regime, behavioral and biochemical evaluation in our experiments. The doses of scopolamine (25 μg/kg, i.p.) referenced the previous studies (Wohleb et al., 2016; Yu et al., 2017, 2018) and the dose of reserpine based on our preliminary experiments.

2.3. Experimental design

The mice were first divided into two groups ($n = 24$ per group), and then treated daily with reserpine (1.5 mg/kg, i.p.) or saline for 10 days. 24 h and 48 h after the last injection, the open field test (OFT) and forced swim test (FST) were performed, on days 11 and 12, respectively. Twenty-four hours after the FST, the saline and reserpine groups were each further divided into two groups, as follows: (1) saline plus saline; (2) saline plus scopolamine; (3) reserpine plus saline; (4) reserpine plus scopolamine. The saline or scopolamine (25 μg/kg, i.p.) was injected daily for 3 days. Twenty-four and 48 h after the last drug treatment, mice were submitted to the OFT and FST on days 16 and 17, respectively.

2.4. Open field test (OFT)

Briefly, the mice were placed in a Plexiglas box (50 × 50 × 39 cm) to observe their reaction to an open field. A digital camera that could cover the entire field was placed above the box. The box was cleaned thoroughly with 1% ethanol before each animal was tested. At the beginning of the test, the animal was placed in the center of the arena and allowed to freely explore for 5 min, and its movement was recorded using the digital camera. During the 5 min of exploration, the numbers of line crossings and rearings were recorded for use in estimating the possible effects of drug treatment on locomotor activity.

2.5. Forced swim test (FST)

In this test, the mice were plunged individually into a clear Plexiglas cylinder (60 cm height, 20 cm diameter) containing approximately 30 cm of water (21 ~ 25 °C) and were allowed to swim for 6 min. Total immobility times (i.e., making only minimal movements to keep the head above water or floating) during the last 4 min of the 6-min test period were recorded.

2.6. Western blotting analysis

After all behavioral tests, the mice were sacrificed by cervical dislocation, and the hippocampus and PFC were rapidly removed from the brain. Then, the whole hippocampus and PFC tissues were lysed in SDS buffer containing protease inhibitors, followed by centrifugation at 13,200 rpm for 30 min. The supernatant was collected, and the protein concentration was determined with a bicinchoninic acid (BCA) assay kit. Equal amounts of protein were separated by 12% SDS-PAGE and transferred to PVDF membranes. After blocking with 5% BSA for 1 h, the membranes were incubated with primary antibodies overnight at 4 °C. Primary antibodies against the following proteins were used: 5-HTT, BDNF, TPH1 and GAPDH. After washing twice in TBST (10 min each), the membrane was treated with HRP-conjugated secondary antibody diluted in TBST for 1 h at room temperature. After washing in TBST 4 times (10 min each), bands were detected and quantified using a fluorescence scanner (Odyssey Infrared Imaging System, LI-COR Biotechnology, Lincoln, NE).

2.7. Statistical analysis

Data are presented as the mean ± standard error of the mean (SEM). Statistical analysis was conducted with two-tailed Student's *t*-tests (for two-group comparisons) and two-way ANOVA (or four-group comparisons) using GraphPad Prism software (Version 5.0, Prism software for PC, GraphPad, USA), and differences were considered significant at $p < 0.05$ or less, where specified.

3. Results

3.1. Scopolamine significantly reversed the depression-like behaviors induced by reserpine in mice

As demonstrated in Fig. 1B, C and D, 10 days of administration of reserpine (1.5 mg/kg, i.p.) significantly decreased the line crossings [$t = 3.727$, $df = 46$, $p = 0.0005$, Fig. 1B] and rearings [$t = 4.571$, $df = 46$, $p < 0.0001$, Fig. 1C] in the OFT and increased the immobility time [$t = 3.701$, $df = 46$, $p < 0.0006$, Fig. 1D] in the FST, showing a depression-like state in mice. In addition, Fig. 1E and F showed that the administration of reserpine significantly affected the line crossings [$F(1, 44) = 28.23$; $p < 0.0001$, Fig. 1E] and rearings [$F(1, 44) = 44.04$; $p < 0.0001$, Fig. 1F] in mice. However, scopolamine id not modify the changes of the line crossing of OFT [$F(1, 44) = 3.812$; $p = 0.0573$] and rearings [$F(1, 44) = 1.412$; $p = 0.2410$] of mice in the OFT, indicating that the antidepressant-like effects of scopolamine were not associated

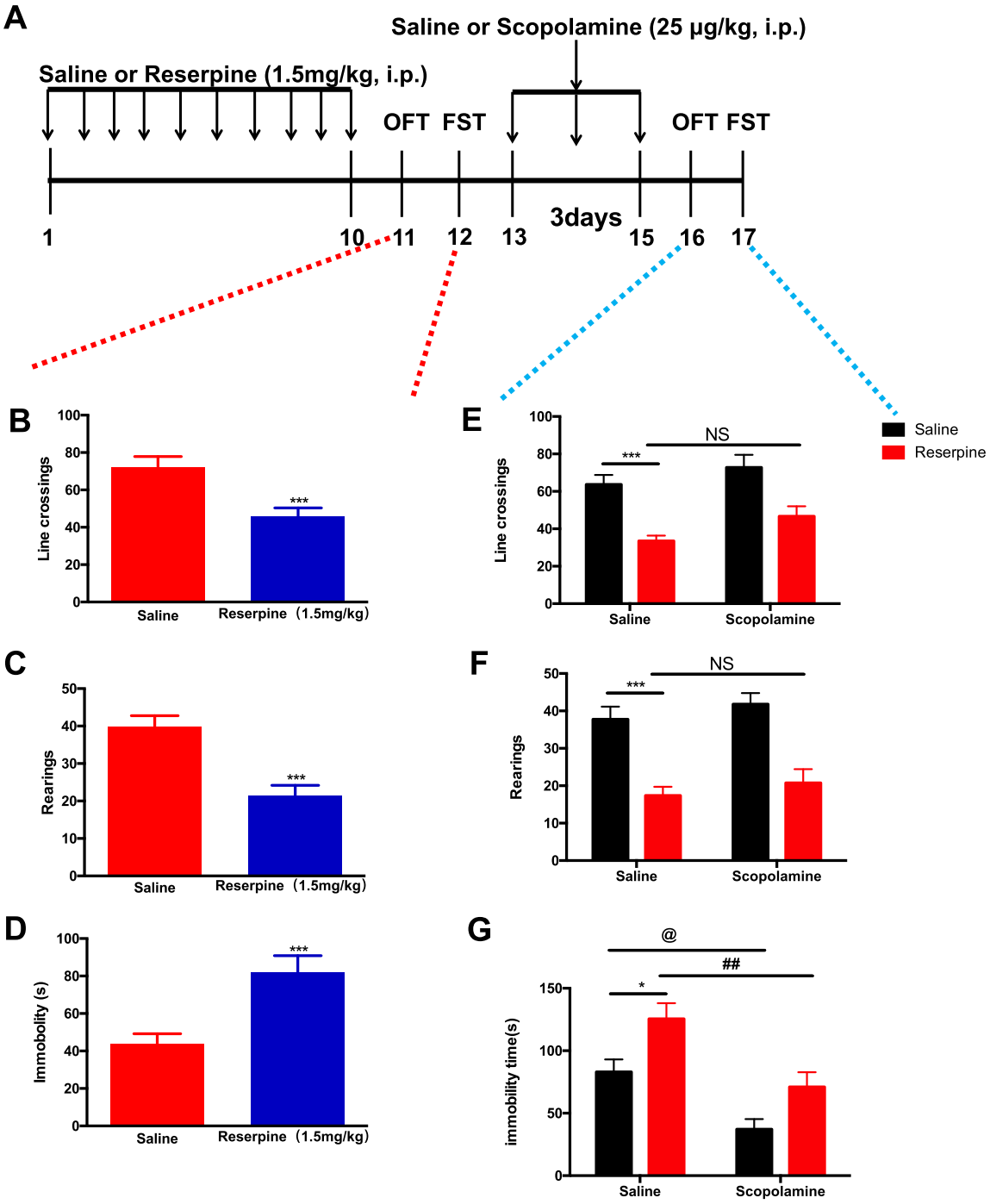

Fig. 1. Scopolamine treatment showed antidepressant-like effects in mice with reserpine-induced depression. (A) Timeline of the experiments. (B-C) Chronic reserpine administration induced a significant decrease in mouse locomotor activity in the OFT. (D) Chronic reserpine administration induced a significant increase in mouse immobility time in the FST. (E-F) Sub-chronic treatment of mice with scopolamine did not reverse the reserpine-induced decrease in line crossings and rearings in the OFT. (G) Scopolamine significantly reversed the reserpine-induced decrease in immobility time in mice. The data shown are the means ± standard error of the mean (SEM). (B-D) $n = 24$ per group, two-tailed Student's *t*-tests, ***p < 0.001 vs. saline group. (E-G) $n = 12$ per group. Two-way ANOVA, $^{*}p < 0.05$ indicates the comparison between the saline + reserpine group and the saline + saline group; $^{\#\#}p < 0.01$ indicates the comparison between the reserpine + saline group and the reserpine + scopolamine group; $^{@}p < 0.05$ indicates the comparison between the saline + scopolamine group and the saline + saline group.

with locomotor activity. Furthermore, to investigate the antidepressant-like effects of scopolamine, the two-way ANOVA revealed significant differences for scopolamine treatment [F (1,44) = 21.96; $p < 0.0001$, Fig. 1G], reserpine treatment [F (1, 44) = 12.69; $p = 0.0009$, Fig. 1G]. However, there was no significant difference for scopolamine treatment × reserpine interaction [F (1,44) = 0.1622; $p = 0.6891$, Fig. 1G]. Post-hoc analysis showed that the increase of the immobility time by reserpine ($p < 0.05$) was significantly decreased by scopolamine treatment ($p < 0.01$).

3.2. Scopolamine significantly reversed the reserpine-induced down-regulation of 5-HTT, TPH1 and BDNF in the mouse hippocampus and PFC

As shown in Fig. 2, the two-way ANOVA revealed significant differences for scopolamine treatment [hippocampus: 5-HTT: F (1,16) = 80.51; $p < 0.0001$, Fig. 1B; BDNF: F (1,16) = 61.23;

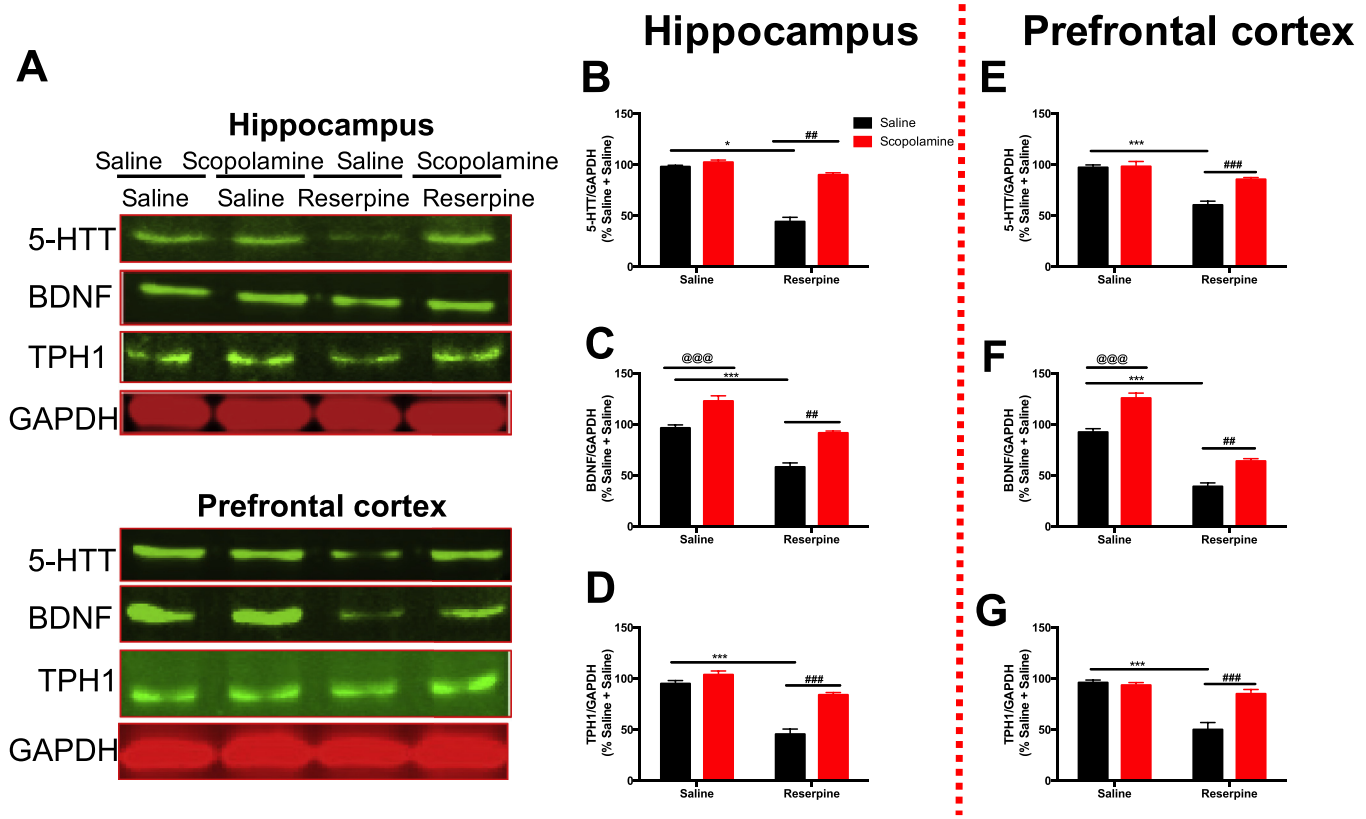

Fig. 2. Scopolamine reversed the down-regulation of 5-HTT, BDNF and TPH1 in the mouse hippocampus and PFC. (A) Representative western blots of 5-HTT, BDNF and TPH1 from the mouse hippocampus and PFC. The remaining panels show the quantification of the immunoblotting bands of 5-HTT (B and E), BDNF (C and F) and TPH1 (D and G) in the hippocampus and PFC respectively. The data are shown as the means ± SEM. $n = 5$ per group. Two-way ANOVA, $^{*}p < 0.05$, $^{***}p < 0.001$ indicates the comparison between the saline + reserpine group and the saline + saline group; $^{\#\#}p < 0.01$, $^{\#\#\#}p < 0.001$ indicates the comparison between the reserpine + saline group and the reserpine + scopolamine group; $^{@@@}p < 0.001$ indicates the comparison between the saline + scopolamine group and the saline + saline group.

$p < 0.0001$, Fig. 1C; TPH1: F (1,16) = 40.14; $p < 0.0001$, Fig. 1D; PFC: 5-HTT: F (1,16) = 13.65; $p = 0.0020$, Fig. 1E; BDNF: F (1,16) = 59.49; $p < 0.0001$, Fig. 1F; TPH1: F (1,16) = 12.61; $p = 0.0027$, Fig. 1G] on the expression of 5-HTT, BDNF and TPH1 in the hippocampus and PFC respectively. Post-hoc analysis showed that the decreases of 5-HTT [hippocampus: $p < 0.01$; PFC: $p < 0.001$], BDNF [hippocampus: $p < 0.01$; PFC: $p < 0.01$] and TPH1 [hippocampus: $p < 0.001$; PFC: $p < 0.001$] by reserpine were significantly reversed by scopolamine in the hippocampus and PFC of mice. Notably, single treatment with scopolamine significantly increased the BDNF levels in the mouse hippocampus ($p < 0.001$) and PFC ($p < 0.001$) compared with the saline plus saline group.

4. Discussion

We demonstrated for the first time that scopolamine ameliorates the depression-like behaviors induced by reserpine in mice. The scopolamine-induced restoration of BDNF, 5-HTT and TPH1 in the hippocampus and PFC may contribute to the inhibition of reserpine-induced depression-like behaviors.

5-HT is a key neurotransmitter that modulates many neuronal functions, and a large body of experimental and clinical data has demonstrated that the depression, as a disease of the central nervous system, may be regulated by the 5-HT system (López-Figueroa et al., al.,2004; Thakare et al., 2017). Reserpine can irreversibly inhibit the vesicular uptake of monoamines, including 5-HT, and injection of reserpine or its metabolites is widely used to induce depressive-like behaviors in rodents (Iritani et al., 2006; Arora et al., 2011; Yu et al., 2015; Li et al., 2016). However, the mechanism of reserpine-induced 5-HT transmission and depression-like behavior dysfunction is unclear. Herein, the behavioral state of the reserpine-treated mice was evaluated based on the OFT and FST. Our results are consistent with previous studies that have shown that chronic treatment with reserpine significantly decreased locomotor activity and increased immobility time in mice, indicating a typical depression-like phenotype. Several studies have reported decreases in 5-HTT density (Leake et al., 1991; Arango et al., 1995) and 5-HTT binding (Joensuu et al., 2007) in various brain regions in depressed patients. In our present study, 5-HTT protein expression, in both the hippocampus and the PFC, was significantly decreased in the reserpine-induced depressive state in mice. In agreement with our findings, decreased hippocampal 5-HTT protein expression in rodents has been induced by unpredictable chronic mild stress (UCMS) (Li et al., 2017), chronic social stress (McKittrick et al., 2000) and corticosterone (CORT) (Tang et al., 2014). Based on a previous study that found that 5-HTT knockout mice show several behavioral changes, including increased anxiety-like behavior, increased sensitivity to stress, and inhibited exploratory locomotion (Holmes et al., 2003), we speculated that 5-HTT downregulation in the mouse hippocampus and PFC may trigger the behavioral dysfunction induced by reserpine.

Several lines of evidence suggest that BDNF is involved in depression, such that the expression of BDNF is decreased in limbic and cortical regions in rodent chronic stress models and in postmortem brains of depressed subjects (Kerling et al., 2017; Zhao et al., 2017; Phillips, 2017). However, whether BDNF was down-regulated by chronic reserpine treatment in mice was unknown. In our current work, we also found that reserpine significantly decreased the BDNF levels in both the hippocampus and PFC in mice. It is well known that 5-HTT and BDNF in the central nervous system play a key role in the pathophysiology of

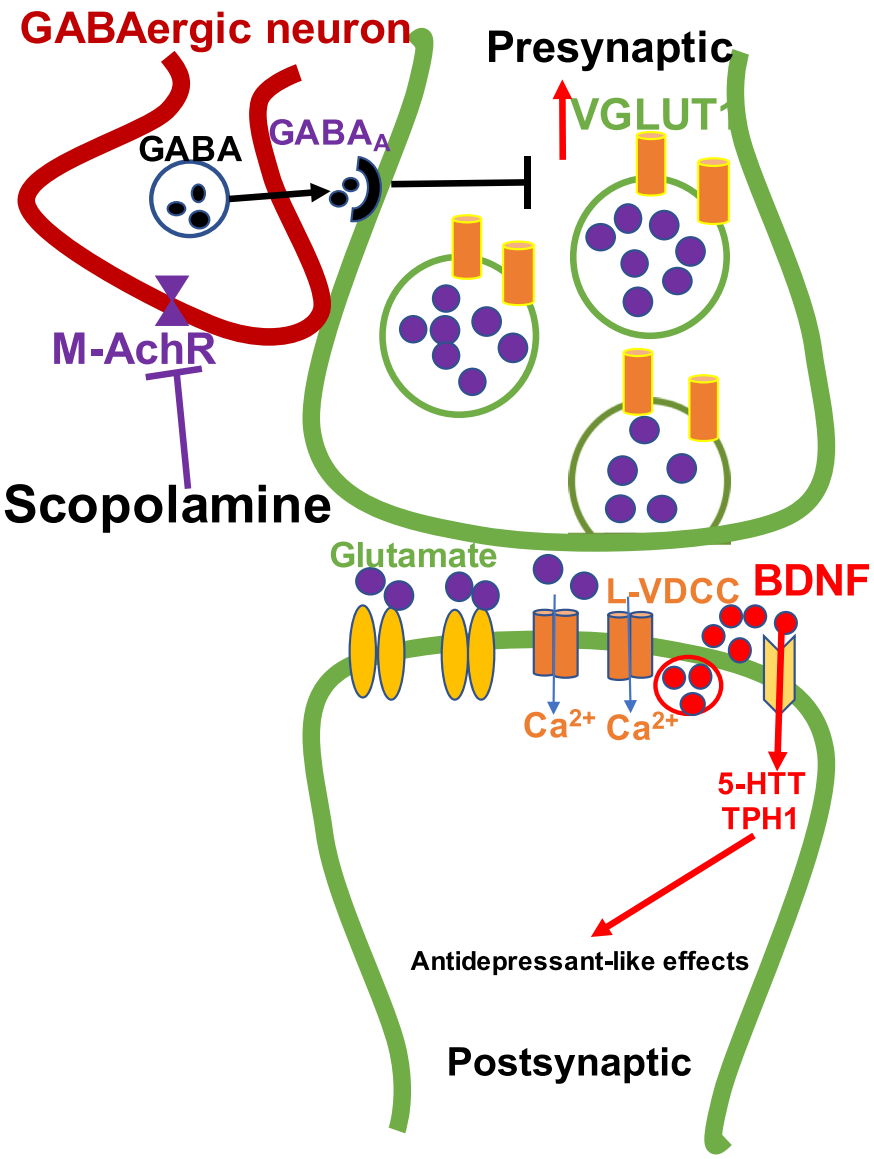

Fig. 3. The may underlying mechanisms of rapid acting antidepressant-like effects of scopolamine in reserpine-induced mice.

depression, and the alterations in the central nervous system are associated with the mechanisms of action underlying the therapeutic activity of antidepressant drugs (Qiu et al., 2013; Scabia et al., 2018). Additionally, recently work identify an essential role for activity-dependent BDNF release in the rapid antidepressant effects of scopolamine (Ghosal et al., 2018). Therefore, reversal of reserpine-induced depression-like behaviors in mice by treatment with scopolamine was performed in the present study to explore effects of scopolamine on the monoamine pathways in the mouse brain. In the present study, the administration of scopolamine (25 μg/kg, i.p., once per day for 3 days) significantly decreased the increase in immobility time of mice in the FST, indicating significant antidepressant-like effects. To the best of our knowledge, the present study is the first to report the antidepressant-like effects of scopolamine in reserpine-induced anhedonia in mice. However, scopolamine produced no changes in the reserpine-induced decrease in locomotor activity in mice. Moreover, scopolamine treatment also significantly elevated the levels of 5-HTT and BDNF in the mouse hippocampus and PFC. Although recent studies have been conducted to explore BDNF release in the antidepressant-like effects of scopolamine in mice (Yu et al., 2017; Ghosal et al., 2018), very little is known about the antidepressant-like mechanism of scopolamine, including whether it acts via reversing the reserpine-induced downregulation of BDNF and then down-regulation of 5-HTT in the mouse hippocampus and PFC. Given that the previous study revealed that the serotonin transporter function is modulated by BDNF (Mössner et al., 2000), further studies should be conducted to explore the regulatory relationship between BDNF and 5-HTT in the antidepressant-like effects of scopolamine in mice with reserpine-induced anhedonia.

Interestingly, TPH1 catalyzes the rate-limiting step of 5-HT biosynthesis in the serotonergic neurons and several works have pointed out that TPH1 gene may be associated with depression and may a possible candidate involved in the etiology of depression (Gizatullin et al., 2006; Jokela et al., 2007). In the current study, we found that the expression of TPH1 was significantly decreased by reserpine in the hippocampus and PFC of mice, which may partly explain the mechanisms of depression-like behaviors induced by reserpine in mice. Notably, we also demonstrated that this TPH1 down-regulation was significantly reversed by scopolamine. Based on the previous studies which reported that BDNF–mediated signaling is crucial in the phenotypic development of serotonergic properties and up-regulation of TPH (Galter and Unsicker, 2000; Eaton et al., 1995), we speculated that the up-regulation on TPH1 of scopolamine may via the BDNF-mediated signaling.

In conclusion, scopolamine exerts antidepressant-like effects on reserpine-induced mouse model and the working model of the potential mechanism of scopolamine was shown in Fig. 3. Our results demonstrated that the antidepressant-like effects of scopolamine are mediated, at least in part, via modulating the 5-HTT and TPH1 by BDNF in the hippocampus and PFC of mice. Scopolamine acts via blockade of M-AchR receptors located on GABAergic interneurons (Navarria et al., 2015; Wohleb et al., 2016), resulting in disinhibition of glutamate neurons and release of glutamate in mice. The increase in glutamate transmission of mice caused depolarization of the neuron and an influx of calcium through L-VDCC (Lepack et al., 2014, 2016; Yu et al., 2017). Additionally, depolarization of the neuron induces rapid release of BDNF, and may via TrkB to regulate the 5-HTT and TPH1 in the actions of scopolamine. The limitation of the current study is whether the reversal effect of scopolamine on 5-HTT and TPH1 dependent on the BDNF-mediated signaling remains unknown. Further investigations are needed to reveal the mechanisms of BDNF down-regulation on the 5-HTT and TPH1 in the antidepressant-like effect of scopolamine in mice with reserpine-induced anhedonia.

Acknowledgments

This research was supported by National Natural Science Foundation of China (No. 81671337; No.81201050), Natural Science Foundation of Zhejiang province (No. LQ12H09001); Natural Science Foundation of Ningbo (No. 2017A610212; No. No.2016A610156); Ningbo Municipal Innovation Team of Life Science and Health (2015C110026); Xinmiao Talents Program of Zhejiang (2018R405052); Student Research and Innovation Program (SIRP);This project is also sponsored by K.C. Wong Magna funded in Ningbo University. The authors declare that they have no competing interests.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.psychres.2018.12.015.

References

Arango, V., Underwood, M.D., Gubbi, A.V., Mann, J.J., 1995. Localized alterations in pre- and postsynaptic serotonin binding sites in the ventrolateral prefrontal cortex of suicide victims. Brain Res. 688, 121–133.

Arora V., KuhadA., Tiwari, V., Chopra, K., 2011. Curcumin ameliorates reserpine-induced pain-depression dyad: behavioural, biochemical, neurochemical and molecular evidences. Psychoneuroendocrinology 36 (10), 1570–1581.

Bregman, T., Nona, C., Volle, J., Diwan, M., Raymond, R., Fletcher, P.J., et al., 2017. Deep brain stimulation induces antidepressant-like effects in serotonin transporter knockout mice. Brain Stimul piiS1935-861X(17)30964-6.

Daniele, A., Divella, R., Paradiso, A., Mattioli, V., Romito, F., Giotta, F., et al., 2011. Serotonin transporter polymorphism in major depressive disorder (MDD), psychiatric disorders, and in MDD in response to stressful life events: causes and treatment with antidepressant. In Vivo 25 (6), 895–901.

Dannlowski, U., Kugel, H., Redlich, R., Halik, A., Schneider, I., Opel, N., et al., 2014. Serotonin transporter gene methylation is associated with hippocampal gray matter volume. Hum. Brain Mapp 35 (11), 5356–5367.

Daws, L.C., Munn, J.L., Valdez, M.F., Frosto-Burke, T., Hensler, J.G., 2007. Serotonin transporter function, but not expression, is dependent on brain-derived neurotrophic factor (BDNF): in vivo studies in BDNF-de"cient mice. J. Neurochem. 101, 641–651.

Dong, J., Zhou, Q., Wei, Z., Yan, S., Sun, F., Cai, X., 2017. Protein kinase A mediates scopolamine-induced mTOR activation and an antidepressant response. J. Affect Disord. 227, 633–642.

Eaton, M.J., Whittemore, S.R., 1996. Autocrine BDNF secretion enhances the survival and serotonergic differentiation of rapheneuronal precursor cells grafted into the adult rat CNS. Exp Neurol. 140 (2), 105–114.

Eaton, M.J., Staley, J.K., Globus, M.Y., Whittemore, S.R., 1995. Developmental regulation of early serotonergic neuronal differentiation: the role of brain-derived neurotrophic factor and membrane depolarization. Dev. Biol. 170 (1), 169–182.

Galter, D., Unsicker, K., 2000. Brain-derived neurotrophic factor and trkB are essential for cAMP-mediated induction of the serotonergic neuronal phenotype. J. Neurosci. Res. 61 (3), 295–301.

Gizatullin, R., Zaboli, G., Jönsson, E.G., Asberg, M., Leopardi, R., 2006. Haplotype analysis reveals tryptophan hydroxylase (TPH) 1 gene variants associated with major depression. Biol. Psychiatry 59 (4), 295–300.

Ghosal, S., Bang, E., Yue, W., Hare, B.D., Lepack, A.E., Girgenti, M.J., et al., 2018. Activity-dependent brain-derived neurotrophic factor release is required for the rapid antidepressant actions of Scopolamine. Biol. Psychiatry 83 (1), 29–37.

Goggi, J., Pullar, I.A., Carney, S.L., Bradford, H.F., 2002. Modulation of neurotransmitter release induced by brain-derived neurotrophic factor in rat brainstriatal slices in vitro. Brain Res. 941 (1-2), 34–42.

Hansson, S.R., Mezey, E., Hoffman, B.J., 1998. Serotonin transporter messenger RNA in the developing rat brain: early expression in serotonergic neurons and transient expression in non-serotonergic neurons. Neuroscience 83 (4), 1185–1201.

Holmes, A., Yang, R.J., Lesch K, P, Crawley J, N, Murphy, D.L., 2003. Mice lacking the serotonin transporter exhibit 5-HT(1A) receptor-mediated abnormalities in tests for anxiety-like behavior. Neuropsychopharmacology 28 (12), 2077–2088.

Iritani, S., Tohgi, M., Arai, T., Ikeda, K., 2006. Immunohistochemical study of the serotonergic neuronal system in an animal model of the mood disorder. Exp. Neurol. 201 (1), 60–65.

Ikram, H., Haleem, D.J., 2017. Repeated treatment with reserpine as a progressive animal model of depression. Pak. J. Pharm. Sci. 30 (3), 897–902.

Joensuu, M., Tolmunen, T., Saarinen, P.I., Tiihonen, J., Kuikka, J., Ahola, P., et al., 2007. Reduced midbrain serotonin transporter availability in drug-naïve patients with depression measured by SERT-specific [(123)I] nor-beta-CIT SPECT imaging. Psychiatry. Res. 154 (2), 125–131.

Jokela, M., Räikkönen, K., Lehtimäki, T., Rontu, R., Keltikangas-Järvinen, L., 2007. Tryptophan hydroxylase 1 gene (TPH1) moderates the influence of social support on depressivesymptoms in adults. J. Affect. Disord. 100 (1-3), 191–197.

Kerling, A., Kück, M., Tegtbur, U., Grams, L., Weber-Spickschen, S., Hanke, A., et al., 2017. Exercise increases serum brain-derived neurotrophic factor in patients with major depressive disorder. J. Affect. Disord. 215, 152–155 2017.

Khnychenko, L.K., Yakovleva, E.E., Bychkov, E.R., Shabanov, P.D., 2017. Effects of fluorencarbonic acid derivative on the levels of monoamines and their metabolites in brain structures of rats with modeled depression-like state. Bull. Exp. Biol. Med. 163 (5), 632–634.

Leake, A., Fairbairn, A.F., McKeith, I.G., Ferrier, I.N., 1991. Studies on the serotonin uptake binding site in major depressive disorder and control post-mortem brain: neurochemical and clinical correlates. Psychiatry. Res. 39 (2), 155–165.

Lepack, A.E., Bang, E., Lee, B., Dwyer, J.M., Duman, R.S., 2016. Fast-acting antidepressants rapidly stimulate ERK signaling and BDNF release in primary neuronal cultures. Neuropharmacology 111, 242–252.

Lepack, A.E., Fuchikami, M., Dwyer, J.M., Banasr, M., Duman, R.S., 2014. BDNF release is required for the behavioral actions of ketamine. Int. J. Neuropsychopharmacol. 18.

Li, Y., Cheng, K.C., Liu, K.F., Peng, W.H., Cheng, J.T., Niu, H.S., 2017. Telmisartan activates PPARδ to improve symptoms of unpredictable chronic mild stress-induced depression in mice. Sci. Rep. 7 (1), 14021.

Li, S., Han, J., Wang, D.S., Feng, B., Deng, Y.T., Wang, X.S., et al., 2016. Echinocystic acid reduces reserpine-induced pain/depression dyad in mice. Metab. Brain Dis. 31 (2), 455–463.

López-Figueroa, A.L., Norton, C.S., López-Figueroa, M.O., Armellini-Dodel, D., Burke, S., Akil, H., et al., 2004. Serotonin 5-HT1A, 5-HT1B, and 5-HT2A receptor mRNA expression in subjects with major depression, bipolar disorder, and schizophrenia. Biol. Psychiatry 55 (3), 225–233.

McKittrick C.R., MagariñosA.M., Blanchard, D.C., Blanchard, R.J., McEwen, B.S., Sakai, R.R., 2000. Chronic social stress reduces dendritic arbors in CA3 of hippocampus and decreases binding to serotonin transporter sites. Synapse 36 (2), 85–94.

Mössner, R., Daniel, S., Albert, D., Heils, A., Okladnova, O., Schmitt, A., et al., 2000. Serotonin transporter function is modulated by brain-derived neurotrophic factor (BDNF) but not nerve growth factor (NGF). Neurochem. Int. 36 (3), 197–202.

Navarria, A., Wohleb, E.S., Voleti, B., Ota, K.T., Dutheil, S., Lepack, A.E., et al., 2015. Rapid antidepressant actions of scopolamine: role of medial prefrontal cortex and M1-subtype muscarinic acetylcholine receptors. Neurobiol. Dis. 82, 254–261.

Newberg, A.B., Amsterdam, J.D., Wintering, N., Shults, J., 2012. Low brain serotonin transporter binding in major depressive disorder. Psychiatry. Res. 202 (2), 161–167.

Park, L, Furey, M, Nugent, AC, Farmer, C, Ellis, J, Szczepanik, J, et al., 2018. Neurophysiological changes associated with antidepressant response to Ketamine not observed in a negative trial of Scopolamine in major depressive disorder. Int. J. Neuropsychopharmacol. https://doi.org/10.1093/ijnp/pyy051. 2018 Sep 1.

Pérez-Olmos, I., Bustamante, D., Ibáñez-Pinilla, M., 2016. Serotonin transporter gene (5-HTT) polymorphism and major depressive disorder in patients in Bogotá, Colombia. Biomedica 36 (2), 285–294.

Phillips, C., 2017. Brain-derived neurotrophic factor, depression, and physical activity: making the neuroplastic connection. Neural. Plast., 7260130.

Qiu, H.M., Yang, J.X., Wu, X.H., Li, N., Liu, D., Wang, L.J., et al., 2013. Antidepressive effect of paroxetine in a rat model: upregulating expression of serotonin and norepinephrine transporter. Neuroreport 24 (10), 520–525.

Rigal, A., Mouchabac, S., Peretti, C.S., 2016. Interest of scopolamine as a treatment of major depressive disorder. Encephale 42 (6), 568–573.

Rumajogee, P., Madeira, A., Vergé, D., Hamon, M., Miquel, M.C., 2002. Up-regulation of the neuronal serotoninergic phenotype in vitro: BDNF and cAMP share Trk B-dependent mechanisms. J. Neurochem. 83 (6), 1525–1528.

Scabia, G., Barone, I., Mainardi, M., Ceccarini, G., Scali, M., Buzzigoli, E., et al., 2018. The antidepressant fluoxetine acts on energy balance and leptin sensitivity via BDNF. Sci. Rep. 8 (1), 1781.

Sivolap, Y.P., 2017. Serotonin neurotrasmission and treatment options for depression. Zh Nevrol Psikhiatr Im S S Korsakova 117 (11), 174–177.

Siuciak, J.A., Clark, M.S., Rind, H.B., Whittemore, S.R., Russo, A.F., 1998. BDNF induction of tryptophan hydroxylase mRNA levels in the rat brain. J. Neurosci. Res. 52 (2), 149–158.

Tang, M., He, T., Sun, X., Meng, Q.Y., Diao, Y., Lei, J.Y., et al., 2014. Subregion-specific decreases in hippocampal serotonin transporter protein expression and function

associated with endophenotypes of depression. Hippocampus 24 (4), 493–501.
Tang, M., Lei, J., Sun, X., Liu, G., Zhao, S., 2013. Stress-induced anhedonia correlates with lower hippocampal serotonin transporter protein expression. Brain Res. 1513, 127–134.
Wohleb, E.S., Wu, M., Gerhard, D.M., Taylor, S.R., Picciotto, M.R., Alreja, M., et al., 2016. GABA interneurons mediate the rapid antidepressant-like effects of scopolamine. J. Clin. Invest. 126, 2482–2494.
Xiao, Q., Pawlyk, A., Tejani-Butt, S.M., 1999. Reserpine modulates serotonin transporter mRNA levels in the rat brain. Life Sci. 64 (1), 63–68.
Yu, H., Li, M., Shen, X., Lv, D., Sun, X., Wang, J., et al., 2018. The requirement of L-type voltage-dependent calcium channel (L-VDCC) in the rapid-acting antidepressant-like effects of scopolamine in mice. Int. J. Neuropsychopharmacol. 21 (2), 175–186.
Yu, H., Li, M., Zhou, D., Lv, D., Liao, Q., Lou, Z., et al., 2017. Vesicular glutamate transporter 1 (VGLUT1)-mediated glutamate release and membrane GluA1 activation is involved in the rapid antidepressant-like effects of scopolamine in mice. Neuropharmacology 131, 209–222.
Yu, H.L., Sun, L.P., Li, M.M., Quan, Z.S., 2015. Involvement of norepinephrine and serotonin system in antidepressant-like effects of oleoylethanolamide in the mice models of behavior despair. Neurosci. Lett. 593, 24–28.
Zhao, M., Chen, L., Yang, J, Han, D., Fang, D., Qiu, X., et al., 2017. BDNF Val66Met polymorphism, life stress and depression: a meta-analysis of gene-environment interaction. J. Affect. Disord. 227, 226–235.