Study on Antidepressant Effect and Mechanism of Crocin Mediated by the mTOR Signaling Pathway

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Abstract
Crocin is a monomer of Chinese traditional herbs extracted from saffron, relieving depression-like behavior. However, its underlying mechanism of action remains unclear. Herein, we explored whether crocin’s antidepressant effect depended on the mammalian target of the rapamycin (mTOR) signaling pathway. The model of PC12 cells injury was established by corticosterone, the changes in cell survival rate were tested by the CCK-8 method, and the changes in cellular morphology were observed under a fluorescence microscope. The depression model was established by chronic unpredictable mild stress (CUMS), and its antidepressant effect was estimated by open field test (OFT), forced swimming test (FST), and tail suspension test (TST). Western blot was used to monitor the protein expression. The results showed that crocin could effectively improve cell survival rate and cellular synaptic growth, alleviate the depressive behavior of CUMS mice, and promote the expression of BDNF, P-mTOR, P-ERK, and PSD95. However, when rapamycin was pretreated, the antidepressant effects of crocin were inhibited. In summary, crocin plays a significant antidepressant effect. After pretreatment with rapamycin, the anti-depression effect of crocin was significantly inhibited. It is suggested that the mechanism of the anti-depression effect of crocin may be related to the mTOR signaling pathway.

Graphical Abstract

Keywords Depression · Crocin · Mouse · Rapamycin · mTOR · Lateral ventricle

Extended author information available on the last page of the article
Abbreviations

CUMS  Chronic unpredictable mild stress  
OFT  Open field test  
FST  Forced swimming test  
TST  Tail suspension test  
mTOR  Mammalian target of rapamycin  
BDNF  Brain-derived neurotrophic factor  
ERK  Extracellular signal-regulated kinase  
PSD95  Postsynaptic density 95  
MDD  Major depressive disorder  
PC12  Pheochromocytoma cell 12  
CORT  Corticosterone  
DMEM  Dulbecco’s modified eagle medium  
FBS  Fetal bovine serum  
SDS-PAGE  Sodium dodecyl sulfate-polyacrylamide gel electrophoresis  
ANOVA  One-way analysis of variance  
M  Model  
VLC  Very low-dose crocin 1 µmol L⁻¹  
LC  Low-dose crocin 10 µmol L⁻¹  
HL  High-dose crocin 30 µmol L⁻¹  
µM  µMol L⁻¹  
F  Fluoxetine 10 µmol L⁻¹  
Rapa  Rapamycin  
TrkB  Tyrosine kinase receptor B  
CREB  CAMP response element-binding protein  
4E-BP1  4E binding protein 1  
S6K1  S6 kinase 1  
FKBP12  FK506-binding protein 12

Introduction

Major depressive disorder (MDD) is a mental disease mainly manifested by depression, pessimism, and functional decline, and its pathogenesis is multiple and persistent. The pathological mechanism of MDD is still unclear, and various causes are often involved. The central nervous system function decline is a significant factor in the onset of MDD. Crocin is a kind of active ingredient extracted from saffron crocus that has a diversiform function in anti-inflammatory, analgesic, neuronal protection, and enhanced memory [1]. In recent years, it has been confirmed that crocin can alleviate neurotoxicity in rotenone-induced Parkinson’s disease rats and improve chronic obstructive pulmonary disease-induced depression by inhibiting PI3K/Akt-mediated inflammation [2, 3]. Crocin-i can also reduce depressive behavior in mice with chronic corticosterone-induced depression by inhibiting hippocampal neuroinflammation (IL-1β) and oxidative stress and alleviate depression-like behavior by modulating the microbial-gut-brain axis in mice with chronic inhibitory stress [4, 5]. Moreover, studies have shown the antidepressant-like effects of crocin by increasing the levels of P-CREB and brain-derived neurotrophic factor (BDNF) and regulating classic antidepressant molecular signaling, like GHGR-PI3K signaling pathway [6, 7]. However, the specific mechanism of crocin action was not clearly demonstrated.

Mammalian target of rapamycin (mTOR), a serine/threonine-protein kinase, and a target of rapamycin and its analogs can be used to regulate cell growth proliferation and protein synthesis [8]. Studies have found that BDNF expression level and activation of the mTOR signaling pathway are essential for rapid-acting antidepressants to promote dendritic growth and increase synaptic protein content [9]. Ketamine, for example, regulated the expression of postsynaptic density protein (PSD95) and Synapsin-1 by activating mTOR and BDNF signaling pathways in mPFC, instantaneously activating downstream 4e binding protein1 (4E-BP1) and p70S6K [10]. In addition, the depressive behavior of CUMS-induced mice is also associated with reduced levels of p-mTOR and p-p70S6K [11]. Consequently, the mTOR signaling pathway was considered crucial for developing new antidepressant drugs. As a specific inhibitor of the mTOR signaling pathway, rapamycin (rapa) has been identified to inhibit the activation of the mTOR signaling pathway, leading to synaptic dysfunction, thereby blocking the effects of rapid-acting antidepressants. What was certain is that crocin had the function of alleviating depressive behavior and regulating the central nervous system [2–4], but whether the antidepressant effect of crocin depends on the mTOR signaling pathway was still not clear. Rapamycin, a blocker of mTOR, was used in advance to reveal whether the mechanism of acute antidepressant action of crocin depends on the mTOR signaling pathway at both cellular and animal levels, thus providing evidence for the development of new rapid antidepressants.

Materials and Methods

Animals

Male ICR mice, 6–8 weeks old, weighing 22 ± 2 g, were obtained from Hunan Slack Jingda Experimental Animal Co., Ltd. SCXK (Xiang) 2019-0004. (Changsha, China). All animals are kept under standard conditions, with a temperature of (23 ± 2) °C and relative humidity (50–65%). The light follows the laws of nature, free drinking, and eating. Acclimatize for 7 days before the experiment, and then group according to body weight. Detailed steps are shown in Figs. 2A, D, 3B. All animal procedures were performed by the Guidelines for the Care and Use of Laboratory Animals approved by the Animal Care Committee of Jiangxi Yichun University.
Drugs and Antibodies

Crocin, purchased from Shanghai Macleans Biochemical Technology Co., Ltd. (C11523747); Rapamycin, purchased from Soleibao Biotechnology Co., Ltd. (112P034); Fluoxetine hydrochloride, purchased from Shanghai Maclean Biochemical Technology Co., Ltd. (C10221539); Corticosterone (CORT) 98% was purchased from Aladdin Biotechnology Co., Ltd. (11530027); CCK-8 kit was purchased from Bridgen Bridge Biology Co., Ltd. (BRI2134) (China).

Antibody P44/42 MAPK (Erk1/2), Anti-BDNF Rabbit pAb, PSD95, and β-actin were purchased from Chengdu Zhengneng Biotechnology Co., Ltd. (KK0409, 20200611, 20200101, JJ1231). (Chengdu, China); p-P44/42 MAPK (Erk1/2), mTOR, and P-mTOR were purchased from Cell Signaling Technology in the United States (28, 19, and 9) (USA).

Delivery Way

Rapamycin and 0.9% NaCl was administered by lateral ventricle injection. Crocin and drinking water were administered by gavage [12].

Modeling Method and Behavioral Tests

CUMS Model

CUMS simulates the social pressure of patients with depression. The experimental program adopts orphan rearing and 4 consecutive weeks of stress stimulation except for the control group, including: (1) water deprivation for 24 h, (2) fasting for 24 h, (3) wet litter for 24 h, (4) noise (> 80 dB) for 3 h, (5) tail-clamping for 30 min, (6) tilts the squirrel cage at 45° for 24 h, (7) swim in ice water for 5 min, (8) turn upside down day and night, (9) light up all night [13, 14]. Perform 1–2 kinds of stimulation every day to ensure that the stimulation is not repeated within 3 days.

Open Field Test (OFT)

The whole experiment process was carried out in a quiet and dark environment. Before the experiment, the mice were placed in the experimental environment to fit in for 1–2 h. After 30 min of administration, the mice were placed in the bottom center of the square arena (50×50×50 cm), and the number of horizontal crawls and the number of uprights of the mice within 5 min were recorded. Clean the inner wall and the bottom of the arena to avoid the smell left by the last mice [15].

The Forced Swimming (FST)

The FST is also one of the most commonly used methods to measure the extent of despair in animals. The mice were placed in a beaker (20×14 cm in diameter) containing 15 cm deep water at 24 ± 1 °C to swim. The cumulative immobility time (s) was recorded for the last 4 min of a 6 min swim test. The mice were supposed to be motionless when they stopped struggling or produced only small limb movements to keep their head floating above water [16].

The Tail Suspension Experiment (TST)

The TST is often used to test the efficacy of preclinical antidepressants. In a soundproof and visually isolated room, the mice were suspended 30 cm from the ground, and the tail of the mice was fixed with tape about 1 cm from the end. Recording the total immobile time of the last 4 min of the 6 min test [16].

Lateral Ventricle Injection Method

The mice were anesthetized with 3% pentobarbital and fixed to a stereotactic brain localization device. When we find the position of the bregma, the anterior fontanelle is the coordinate point, move forward 0.3–0.6 mm, and move laterally 1 mm to locate [17]. Drill a hole with a bone drill, insert a 1.7 mm microsyringe, and inject 2 μL Trypan blue. Wait 1 h after the injection, and open the brain tissue. If the blue diffuses to other ventricles, the location is successful. The buried pipe repeats the above process, and the customized casing and casing cap are embedded in the borehole. Post-operative observation 1–2 days, if there is no discomfort, recovery 5–7 days, and then the experiment. Rapamycin can be injected directly through the casing. The specific steps are shown in Fig. 3B.

Cell Culture, Modeling, and Treatment

Rat adrenal pheochromocytoma cell 12 (PC12) was purchased from Shanghai Tairan Biotechnology Co., Ltd (Shanghai, China). They were cultured in Dulbecco’s Modified Eagle Medium (DMEM, Gibco, 1645799, USA) with 10% fetal bovine serum (FBS, 4276071k, Gibco) and maintained at an atmosphere composed of 5% CO₂, 37 °C for 24 h (Thermo Fisher Scientific, 311, USA).

After the cells were in the logarithmic growth phase, they were seeded in a 96-well plate with 1 × 10⁴ cell mL⁻¹. After the cells adhered to the wall, different concentrations of CORT (100, 200, 300, 400, and 500 µmol L⁻¹) were added,
continued to culture for 24 h, then measured the cell survival rate by the CCK-8 method. We selected the CORT concentration at which 50% of the cells were damaged for subsequent experiments. The drug administration sequence was as follows: crocin was pretreated for 2 h, then corticosterone was treated for 24 h, 10 μmol L⁻¹ CCK-8 was added into each well for further culture for 1–4 h, and the OD value was determined at 450 nm with a microplate meter. Cell survival rate (%) = [OD (experimental group) − OD (zeroing group)] / OD (control group) × 100%. The synaptic growth was observed and photographed under an inverted fluorescence microscope (×20).

Western Blot

The prefrontal cortex was lysed in RIPA buffer containing protease inhibitors and phosphatase inhibitors. The protein concentration was quantified by Bradford’s method. The protein concentration was measured by BSA, diluted to a uniform concentration, and boiled in boiling water at 100 °C for 5 min. The sample was added to sodium dodecyl sulfate-polyacrylamide gel for electrophoresis separation (SDS-PAGE), and then the protein was transferred to the PVDF membrane with a wet transfer buffer and blocked in a 5% skimmed milk powder solution in TBST 1–1.5 h, the membrane was incubated with mTOR (1:1000), p-mTOR (1:1000), BDNF (1:1000), PSD95 (1:1000), ERK (1:1000), P-ERK (1:1000). Detection was performed with an Alpha Fluor Chem gel imager, and data were analyzed using Image J software. Each experiment was performed more than three times [2].

Statistical Analysis

All data were expressed as mean ± SEM and graphed using the GraphPad Prism 8.0.2 (San Diego, USA). IBM SPSS Statistics 26.0 was used for data analysis. One-way analysis of variance (ANOVA) was used when the data conformed to the normal distribution. The non-parametric test was used when the data did not conform to the normal distribution. P < 0.05 is considered statistically significant.

Results

Crocin had a Significant Effect on Protecting the Survival Rate of Damaged Cells and the Growth of Cell Synapses

The results revealed that PC12 cells were damaged in a concentration-dependent manner in response to different concentrations of corticosterone. When the concentration was 500 μmol L⁻¹, the cell survival rate was about 50% (Fig. 1A; #P < 0.001), which could be used for subsequent modeling. Before corticosterone was administered, crocin was pre-treated for 2 h. The results showed that compared with the model group, all crocin concentrations could significantly improve cell survival rate and promote the growth of cellular synapses (Fig. 1B–D). Among them, 1 μmol L⁻¹ crocin can significantly improve cell survival rate (*P < 0.05) and promote the growth of cellular neurons (**P < 0.01). When the concentration of crocin increased to 10 μmol L⁻¹ and 30 μmol L⁻¹, the effect was more significant (**P < 0.001).

Effects of Crocin on Behavior and Protein Expression in Normal and CUMS-Induced Mice

Crocin was administered to normal mice by gavage, and the behavior was detected after 30 min of drug administration, as shown in Fig. 2A. The results showed that, compared with the model group, the cumulative immobility time of mice in FST and TST was significantly lower after administration of crocin, and the difference was statistically significant (Fig. 2B, C, **P < 0.01, *P < 0.05). In CUMS mice (Fig. 2D), the body weight of mice and the number of crossing and rearing of mice in OFT showed no difference On day 0. (Fig. 2E–G). After the 21 days of modeling, compared with the control group, the weight of the CUMS mice was reduced (Fig. 2E, #P < 0.05, ##P < 0.01), and the number of crossing and rearing in OFT were significantly decreased (Fig. 2F, G, *P < 0.05). Chronic treatment with 10 mg kg⁻¹ and 30 mg kg⁻¹ crocin for 14 days could significantly reversed the CUMS-induced reduction in body weight (Fig. 2E, **P < 0.01, *P < 0.05), and the number of crossing and rearing increased significantly in the high-dose crocin group (Fig. 2F, G, ***P < 0.01, *P < 0.05). In addition, after the 14th day of administration, we also tested behavioral changes in mice during FST and TST. The results showed that compared with the control group, the accumulated immobility time in FST and TST was significantly increased in the model group (Fig. 2H, I, *P < 0.05). Compared with the model group, the accumulated immobility time of mice in the high-dose crocin group was significantly reduced in FST and TST, and the difference was statistically significant (Fig. 2H, I, **P < 0.01, *P < 0.05).

The above results have proved that continuous administration of crocin could effectively relieve depressive behavior in CUMS mice. In order to further determine the antidepressant pathway of crocin, we detected the expression levels of BDNF, P-mTOR/mTOR, P-ERK/ERK, and synaptic protein PSD95 in the prefrontal cortex of mice in each group. Experimental studies found that compared with normal mice, the expression of BDNF, P-mTOR/mTOR, P-ERK/ERK, and PSD95 in the prefrontal cortex of CUMS mice was significantly inhibited (Fig. 2J–M, ###P < 0.001, *P < 0.05). After
Fig. 1 Protective effect of crocin on PC12 cells. A Damage of different concentrations of corticosterone on PC12 cells. (*P < 0.05, **P < 0.01, ***P < 0.001, compared with the control group). B Effects of crocin on the survival rate of corticosterone-induced PC12 cells. (###P < 0.001, compared with the control group; ***P < 0.001, compared with the model group). C, D Effects of crocin on neurons of corticosterone-induced PC12 cells. (**P < 0.01, ***P < 0.001, compared with the model group). One-way ANOVA, (n = 3–4/group)
Fig. 2 The antidepressant effect and molecular expression of crocin. A Procedures of training, drug treatment, and testing in acute depression experiment. FST forced swimming test; TST tail suspension test. B, C Immobility times in the FST and TST 30 min later after acute crocin administration. (***P < 0.001, *P < 0.05, compared with the model group). D Procedures of training, drug treatment, and testing in CUMS. OPT open field test; CUMS chronic mild and unpredictable stress. E Body weight. (†P < 0.05, ††P < 0.01, compared with the control group; **P < 0.01, *P < 0.05, compared with the model group. All comparisons were made within the same time period.). F, G Total crossing number and rearing number in OPT. (²P < 0.05, ²²P < 0.01, compared with the control group; **P < 0.01, *P < 0.05, compared with the model group. All comparisons were made within the same time period.). H, I Immobility times of CUMS mice in the FST and TST. (³P < 0.05, compared with the control group; **P < 0.01, *P < 0.05, compared with the model group). J–M Expression of P-mTOR/mTOR, BDNF, P-ERK/ERK and synaptic proteins PSD95. (⁴P < 0.05, ⁴⁴P < 0.001, compared with the control group; *P < 0.05, **P < 0.01, compared with the model group). One-way ANOVA. (Animal experiment, n = 7–8/group; WB, n = 3–4/group)
14 days of administration, the expression of these proteins increased significantly (Fig. 2J–M, **P < 0.01, *P < 0.05).

**mTOR Plays a Major Role in the Rapid-Acting Antidepressant-Like Process of Crocin**

In order to further determine the role of mTOR in the rapid-acting antidepressant of crocin, we also independently used rapamycin (an antagonist of mTOR) in combination with an effective antidepressant dose of crocin. We gave 0.1 nmol L⁻¹ rapamycin in advance to incubate for 30 min. We then selected the concentration of crocin that was confirmed by the above experiment to significantly improve the cell survival rate (10 μmol L⁻¹, 30 μmol L⁻¹) for co-incubation (Fig. 3A). The results showed that compared with the model group, the cell survival rate of the crocin group was significantly increased (*P < 0.05, ***P < 0.001), but compared with the same concentration of the crocin group, the cell survival rate of the rapamycin pretreatment group was significantly decreased (**P < 0.01). Similarly, rapamycin was administered to the lateral ventricle 30 min in advance in animal models, and the treatment process is shown in the Fig. 3B. The results showed that in the OFT, there was no significant difference in the crossing number and rearing times among each group (Fig. 3C, D). Interestingly, we found that compared with the same concentration of the crocin group, the pretreatment of rapamycin significantly prolonged the cumulative immobility time of mice in FST (Fig. 3E, **P < 0.01), and also reversed the protective effect of crocin on the behavior of depressed mice. Subsequently, we measured the expression of BDNF, P-mTOR/mTOR, P-ERK/ERK, and synaptic protein PSD95 in the prefrontal cortex of mice among each group (Fig. 3F–I). We found that crocin increased the expression of these proteins (**P < 0.001, *P < 0.05, **P < 0.01), but this effect could be significantly blocked by rapamycin (**P < 0.01).

**Discussion**

Before 2020, depression was already one of the main factors causing the global health burden. The global spread of COVID-19 has further aggravated people’s mental health problems and caused the global prevalence of major depression to 27.6% [18]. However, the pathogenesis of depression is still unclear, and existing drugs consist of many problems such as the slow onset of action, many adverse reactions, and significant individual differences [19, 20]. Hence, it is urgent to find new and rapid-acting antidepressants. Studies have shown that crocin played a significant effect in relieving symptoms of depression and ameliorating neuronal damage [2–4]. PC12 cells were derived from a pheochromocytoma cell line of rat adrenal medulla, and it is often used for in vitro studies of neurological diseases.
CORT-treated PC12 cells are also often used to evaluate the effectiveness of antidepressant drugs [21–24]. We investigated the effects of crocin on the survival rate and neurons of CORT-induced PC12 cells. The results suggested that a single administration of crocin could improve the survival rate of PC12 injured cells and ameliorate the morphology of cellular neurons. Then we established the CUMS model and detected the behavioral changes of mice through OFT, FST, and TST. Our results showed that crocin improved the autonomy and exploratory behavior of mice in a new environment, reduced the cumulative immobility time of FST and TST, and up-regulated the expression level of BDNF, PSD95, P-mTOR, and P-ERK in CUMS mice. It further confirmed that crocin has a significant antidepressant effect. Intraventricular administration can directly pass through the blood–brain barrier and take effect quickly, so it is often used as a method of administration to study the mechanism of action of drugs for mental disorders [25, 26]. The intraventricular application of rapamycin quickly blocks the mTOR signaling pathway and inhibits the expression of downstream synaptic proteins. In order to further study the neuroprotective mechanism of crocin, we injected rapamycin into the lateral ventricle to quickly block the mTOR signaling pathway and then gavage crocin 30 mg kg\(^{-1}\) 30 min later. Interestingly, we found that after pretreatment with rapamycin, the neurological effects of crocin and the improvement of depression behavior in mice were significantly inhibited, and the expression of proteins such as BDNF was also significantly reduced. In summary, we believe that the rapid-acting antidepressant of crocin may be mediated by regulating the mTOR signal.

BDNF is considered one of the most widely distributed neurotrophic factors in the central nervous system. It promotes the survival and differentiation of 5-HT neurons and...
plays a crucial role in adult brain neurons’ function and synaptic plasticity [27, 28]. Similarly, BDNF is commonly used to regulate emotions, such as depression and anxiety. Studies have found that the activation of BDNF and its receptor tyrosine kinase receptor B (TrkB) positively stimulated the expression of its downstream signaling pathways, such as mitogen-activated protein kinase (MAPK) and extracellular information-regulated kinase (ERK) [29]. More and more studies were employed to reveal that the treatment of antidepressants can increase the expression level of BDNF in brain tissue and reduce the stress response [30, 31]. In this experiment, it was found that the expression level of BDNF in stressed mice and injured cells was significantly reduced but remarkably increased after administration of crocin, and this effect could be blocked by rapamycin. In addition, BDNF is one of the most widely studied factors that have been identified to be capable of activating ERKs in the hippocampus. ERK further activates a series of protein signaling cascades, such as cAMP response element-binding protein (CREB), which is responsible for regulating neuronal plasticity and memory function [32, 33]. Studies have indicated that crocin exerts neuronal protective effects by stimulating the P-CREB-BDNF signaling pathway [6]. Therefore, we have reasons to believe that the ERK signaling pathway may be one of the targets of crocin action.

mTOR is a signaling pathway downstream of ERK, promoting the phosphorylation of eukaryotic initiation factor 4E-BP1 and S6 kinase 1 (S6K1) to regulate protein synthesis and further stimulate the regeneration of central nervous system neurons and peripheral nerve axes [8, 34]. Studies have identified that ketamine acts quickly by activating the mTOR signaling pathway and then increasing synaptic signaling proteins [35], so mTOR is often regarded as an essential target of rapid antidepressants. As a specific inhibitor of mTOR, rapamycin can bind to the intracellular receptor (FKBP-12) to form a complex to inhibit protein activity [8]. Meanwhile, PSD95, a synaptic protein downstream of mTOR, maintains synaptic plasticity. It plays a crucial role in learning, cerebral ischemia, and oxidative stress. Hence, we hypothesized that crocin targets the BDNF-ERK-mTOR signaling pathway to improve depressive behavior and regulate synaptic protein plasticity. In this study, rapamycin intervention was conducted to investigate the effect of the mTOR signaling pathway on crocin efficacy. Our results also confirmed the hypothesis that a single dose of crocin repaired the damage to cellular neurons, improved depressive behavior, up-regulated the expression levels of BDNF, P-ERK, and P-mTOR, and synaptic protein PSD95 in mice. At the same time, all the above effects were reversed after pretreatment with rapamycin. In conclusion, the antidepressant effect of crocin may be mediated by the mTOR signaling pathway. However, since depression is a complex mental disorder, the antidepressant mechanism of crocin needs to be further determined from various aspects.
Conclusion

As a natural drug component, crocin has rapid-acting antidepressant effects. Its mechanism depends on the activation of mTOR and ERK and the promotion of BDNF and synaptic protein PSD95 expression levels. It provides ideas and a theoretical basis for the clinical development of novel rapid antidepressant drugs.

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Author Contributions CC conceived and designed experiments and manuscript revisions. YW contributed to the design of experiments, analyzed the results, and wrote manuscripts. SZ, XS, SD, BW, and JW performed the experiments. All authors gave final approval for publication.

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Data Availability The data used to support the findings of this study are available from the corresponding author upon request.

Declarations

Conflict of interest The authors have declared that no competing interest exists.

References

1. Manochkumar J, Doss CGP, El-Seedi HR, Efferth T, Ramamooorthy S (2021) The neuroprotective potential of carotenoids in vitro and in vivo. Phytomedicine 91:153676. https://doi.org/10.1016/j.phymed.2021.153676
2. Salama RM, Abdel-Latif GA, Abbas SS, El Magdoub HM, Schaalman MF (2020) Neuroprotective effect of crocin against rotenone-induced Parkinson’s disease in rats: interplay between PI3K/Akt/mTOR signaling pathway and enhanced expression of miRNA-7 and miRNA-221. Neuropharmacology 164:107900. https://doi.org/10.1016/j.neuropharm.2019.107900
3. Xie Y, He Q, Chen H, Lin Z, Xu Y, Yang C (2019) Crocin ameliorates chronic obstructive pulmonary disease-induced depression via PI3K/Akt mediated suppression of inflammation. Eur J Pharmacol. https://doi.org/10.1016/j.ejphar.2019.172640
4. Xiao Q, Shu R, Wu C et al (2020) Crocin-4 alleviates the depression-like behaviors probably via modulating “microbiota-gut-brain” axis in mice exposed to chronic restraint stress. J Affect Disord 276:476–486. https://doi.org/10.1016/j.jad.2020.07.041
5. Xiao Q, Xiong Z, Yu C et al (2019) Antidepressant activity of crocin-I is associated with amelioration of neuroinflammation and attenuation oxidative damage induced by corticosterone in mice. Physiol Behav 212:112699. https://doi.org/10.1016/j.physbeh.2019.112699
6. Moazzafari S, Yasuj SR, Motaghinejad M, Motevalian M, Kheiri R (2019) Crocin acting as a neuroprotective agent against methamphetamine-induced neurodegeneration via CREB-BDNF signaling pathway. Iran J Pharm Res 18(2):745–758. https://doi.org/10.22037/jippr.2019.2393
7. Wu R, Xiao D, Shan X, Dong Y, Tao WW (2020) Rapid and prolonged antidepressant-like effect of crocin is associated with GHSR-mediated hippocampal plasticity-related proteins in mice exposed to prenatal stress. ACS Chem Neurosci. https://doi.org/10.1021/acscchemneuro.c000022
8. Hay N, Sonenberg N (2004) Upstream and downstream of mTOR. Genes Dev 18(16):1926–1945. https://doi.org/10.1101/gad.1212704
9. Park SW, Lee JG, Seo MK et al (2014) Differential effects of antidepressant drugs on mTOR signalling in rat hippocampal neurons. Int J Neuropsychopharmacol 17(11):1831–1846. https://doi.org/10.1017/S1461145714000534
10. Kato T, Pothula S, Liu RJ et al (2019) Sestrin modulator NV-5138 produces rapid antidepressant effects via direct mTORC1 activation. J Clin Invest 129(6):2542–2554. https://doi.org/10.1172/JCI126859
11. Agata C, Iyo AH, Jernigan CS, Beata L, Austin MC, Beata K (2013) Reduced phosphorylation of the mTOR signaling pathway components in the amygdala of rats exposed to chronic stress. Prog Neuropsychopharmacol Biol Psychiatry 40:240–245. https://doi.org/10.1016/j.pnpbp.2012.08.001
12. Tang J, Lu L, Wang Q et al (2020) Crocin reverses depression-like behavior in Parkinson disease mice via VTA-mPFC pathway. Mol Neurobiol 57(7):3158–3170. https://doi.org/10.1007/s12035-020-01941-2
13. Ruiilian L, Honglin Q, Jun X et al (2021) H2S-mediated aerobic exercise antagonizes the hippocampal inflammatory response in CUMS-depressed mice. J Affect Disord 283:410–419. https://doi.org/10.1016/j.jad.2021.02.005
14. Song X, Wang W, Ding S, Liu X, Wang Y, Ma H (2021) Puerarin ameliorates depression-like behaviors of with chronic unpredictable mild stress mice by remodeling their gut microbiota. J Affect Disord 290(April):353–363. https://doi.org/10.1016/j.jad.2021.04.037
15. Gao X, Liang M, Fang Y et al (2018) Deciphering the differential effective and toxic responses of Buprelium Radix following the induction of chronic unpredictable mild stress and in healthy rats based on serum metabolic profiles. Front Pharmacol 8:1–22. https://doi.org/10.3389/fphar.2017.00995
16. Huang X, Li W, You B et al (2020) Serum metabolic study on the antidepressant-like effects of ellagic acid in a chronic unpredictable mild stress-induced mouse model. J Agric Food Chem 68(35):9546–9556. https://doi.org/10.1021/acs.jafc.0c02895
17. Lu C, Liu X, Li J et al (2021) Merazin hydrate produces rapid antidepressant effects depending on activating mTOR signaling by upregulating downstream synaptic proteins in the hippocampus. ACS Chem Neurosci 12(20):3939–3946. https://doi.org/10.1021/acschemneuro.1c00546
18. Santomauro DF, Mantilla Herrera AM, Shadid J et al (2021) Global prevalence and burden of depressive and anxiety disorders in 204 countries and territories in 2020 due to the COVID-19 pandemic. Lancet 390(10132):1700–1712. https://doi.org/10.1016/S0140-6736(21)02143-7
19. Guiard B, Lanfumey L, Gardier A (2006) Microdialysis approach to study serotonin outflow in mice following selective serotonin reuptake inhibitors and substance P (neurokinin 1) receptor antagonist administration: a review. Curr Drug Targets 7(2):187–201. https://doi.org/10.2174/13894500675515428
20. Gahr M, Zeiss R, Lang D, Connemann BJ, Hiemke C, Schönfeldt-Lecuona C (2016) Drug-induced liver injury associated with...
antidepressive psychopharmacotherapy: an explorative assessment based on quantitative signal detection using different MedDRA terms. J Clin Pharmacol 56(6):769–778. https://doi.org/10.1002/jcph.662

21. Lin R, Liu L, Silva M et al (2021) Hederaagenin protects PC12 cells against corticosterone-induced injury by the activation of the PI3K/AKT pathway. Front Pharmacol 12(October):1–13. https://doi.org/10.3389/fphar.2021.712876

22. Zeng B, Li Y, Niu B et al (2016) Involvement of PI3K/Akt/FoxO3a and PKA/CREB signaling pathways in the protective effect of fluoxetine against corticosterone-induced cytotoxicity in PC12 cells. J Mol Neurosci 59(4):567–578. https://doi.org/10.1007/s12031-016-0779-7

23. Tian JS, Wu WZ, Liu SB et al (2021) Stable isotope-resolved metabolomics studies on corticosteroid-induced PC12 cells: a strategy for evaluating glucose catabolism in an in vitro model of depression. J Proteome Res. https://doi.org/10.1021/acs.jproteome.1c00516

24. Yun HY, Jeong Y (2020) Sedum takesimense protects pc12 cells against corticosterone-induced neurotoxicity by inhibiting neural apoptosis. Nutrients 12(12):1–16. https://doi.org/10.3390/nu12123713

25. Taljyan R, Kakoty V, Kc S, Kumari S (2021) Fibroblast growth factor 21 and autophagy modulation ameliorates amyloid β-induced alzheimer disease pathology in rats. Alzheimers Dement 17:e058695. https://doi.org/10.1002/alz.058695

26. Cunha MP, Budni J, Ludka FK et al (2016) Involvement of PI3K/Akt signaling pathway and its downstream intracellular targets in the antidepressant-like effect of creatine. Mol Neurobiol 53(5):2954–2968. https://doi.org/10.1007/s12035-015-9192-4

27. Martinowich K, Lu B (2008) Interaction between BDNF and serotonin: role in mood disorders. Neuropsychopharmacology 33(1):73–83. https://doi.org/10.1038/j.npp.1301571

28. Scharfman H, Goodman J, Macleod A, Phani S, Antonelli C, Croll S (2005) Increased neurogenesis and the ectopic granule cells after intrahippocampal BDNF infusion in adult rats. Exp Neurol 192(2):348–356. https://doi.org/10.1016/j.expneurol.2004.11.016

29. Li C, Sui C, Wang W et al (2021) Baicalin attenuates oxygen-glucose deprivation/reoxygenation–induced injury by modulating the BDNF-TrkB/PI3K/Akt and MAPK/Erk1/2 signaling axes in neuron–astrocyte cocultures. Front Pharmacol 12(June):1–14. https://doi.org/10.3389/fphar.2021.599543

30. Huang Z, Zhong XM, Li ZY, Feng CR, Pan AJ, Mao QQ (2011) Curcumin reverses corticosterone-induced depressive-like behavior and decrease in brain BDNF levels in rats. Neurosci Lett 493(3):145–148. https://doi.org/10.1016/j.neulet.2011.02.030

31. Tsai SJ, Hong CJ, Liou YJ (2010) Effects of bdnf polymorphisms on antidepressant action. Psychiatry Investig 7(4):236–242. https://doi.org/10.4306/pi.2010.7.4.236

32. Sweatt JD (2001) The neuronal MAP kinase cascade: a biochemical signal integration system subserving synaptic plasticity and memory. J Neurochem 76(1):1–10

33. González-Gutiérrez A, Lazo OM, Bronfman FC (2020) The Rab5-rab11 endosomal pathway is required for bdnf-induced CREB transcriptional regulation in hippocampal neurons. J Neurosci 40(42):8042–8054. https://doi.org/10.1523/JNEUROSCI.2063-19.2020

34. Abe N, Borson SH, Gambello MJ, Wang F, Cavalli V (2010) Mammalian target of rapamycin (mTOR) activation increases axonal growth capacity of injured peripheral nerves. J Biol Chem 285(36):28034–28043. https://doi.org/10.1074/jbc.M110.125336

35. Li N, Lee B, Liu RJ, Banasr M, Dwyer JM, Iwata M, Li XY, Aghajanian G, Duman RS (2010) mTOR-dependent synapse formation underlies the rapid antidepressant effects of NMDA antagonists. Science 329(5994):959–964. https://doi.org/10.1126/science.1190287

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