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Chronic pre-treatment with memantine prevents amyloid-beta protein-mediated long-term potentiation disruption★

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Abstract
Previous studies indicate that memantine, a low-affinity N-methyl-D-aspartate receptor antagonist, exerted acute protective effects against amyloid-β protein-induced neurotoxicity. In the present study, the chronic effects and mechanisms of memantine were investigated further using electrophysiological methods. The results showed that 7-day intraperitoneal application of memantine, at doses of 5 mg/kg or 20 mg/kg, did not alter hippocampal long-term potentiation induction in rats, while 40 mg/kg memantine presented potent long-term potentiation inhibition. Then further *in vitro* studys were carried out in 5 mg/kg and 20 mg/kg memantine treated rats. We found that 20 mg/kg memantine attenuated the potent long-term potentiation inhibition caused by exposure to amyloid-β protein in the dentate gyrus *in vitro*. These findings are the first to demonstrate the antagonizing effect of long-term systematic treatment of memantine against amyloid-β protein triggered long-term potentiation inhibition to improve synaptic plasticity.

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Key Words
neural regeneration; neurodegenerative diseases; memantine; amyloid-β protein; long-term potentiation; synaptic plasticity; N-methyl-D-aspartate receptor; Alzheimer's disease; hippocampus; grants-supported paper; neuroregeneration

Research Highlights
(1) Amyloid-β protein inhibited hippocampal long-term potentiation induction in rats.
(2) Low doses of memantine (5 mg/kg or 20 mg/kg) did not alter long-term potentiation induction, but a high dose memantine (40 mg/kg) inhibited hippocampal long-term potentiation induction.
(3) Long-term intraperitoneal injection of 20 mg/kg memantine can rescue amyloid-β protein-mediated inhibition of long-term potentiation induction.

Abbreviations
AD, Alzheimer's disease; Aβ, amyloid-beta protein; LTP, long-term potentiation; NMDAR, N-methyl-D-aspartate receptor; HFS, high frequency stimulation

INTRODUCTION

Alzheimer's disease (AD), the most common cause of senile dementia, is featured by the presence of senile plaques composed of deposits of amyloid-β protein (Aβ), a cleavage product of Aβ precursor protein[1]. Growing evidence indicates that soluble Aβ oligomers may play a more important role in cognitive impairment and neurodegenerative progress in AD than fibrillar Aβ[2]. Previous studies suggested that soluble Aβ impaired synaptic plasticity and resulted in learning and memory deficits, which may occur in early stage AD before irreversible neuronal degeneration[3-5]. Synthetic or cell-derived Aβ solutions have been shown to inhibit the induction of hippocampal long-term potentiation (LTP), *in vivo* [2] and *in vitro* [6]. It has been reported that strategies of interfering with Aβ aggregation or facilitating Aβ clearance have beneficial effects on memory tasks in AD human and animal subjects[7-9], suggesting that soluble Aβ-induced neurotoxicity is involved in the memory deficit observed in the early stages of AD. Blockade of the noxious effect of Aβ on memory could be a potential treatment for alleviating AD symptoms.

Various factors have been reported to be associated with AD, including N-methyl-D-aspartate receptor (NMDAR)-mediated glutamate excitotoxicity. It has been suggested that enhanced glutamatergic neurotransmission by overactivating NMDARs relates to the cognitive deficits observed in AD[10-11]. Many high affinity NMDAR antagonists fail in clinical application because of severe side effects[10]. Memantine is a specific and noncompetitive antagonist of NMDARs and has been approved for the treatment of moderate to severe AD in Europe and the USA. Memantine, characterized by rapid blocking/unblocking kinetics and low-binding affinity, has been postulated to decrease excessive glutamatergic stimulation while at least partially allowing NMDAR physiological activities[12-13]. Memantine has been reported to substantially improve the cognitive function of patients diagnosed with AD[14]. While it remains unknown whether memantine protects plaques from forming, memantine may be neuroprotective against the toxic effects of plaques since Aβ-triggered memory impairment and neuronal toxicity can be relieved by memantine treatment[15-16]. Memantine alleviates neurotoxicity triggered by glutamate or amphetamine derivatives and reduces ischemia-induced neuronal death[17-19]. In support of this hypothesis, recent evidence has been presented that memantine may facilitate ameliorated scopolamine-induced amnesia in day-old chicks (*Gallus gallus domesticus*), and reduce the activity of NMDARs[20]. In addition, memantine increases the durability of synaptic plasticity in moderately aged rats and prevents the impaired LTP resulting from exogenous NMDA or soluble Aβ[21-23]. The mechanisms underlying the neuroprotective effects of memantine in cognitive dysfunction are, however, not fully clarified.

Previous studies have reported that acute treatment with clinically relevant NMDAR blocking doses of memantine attenuated the rapid disruption of hippocampal LTP *in vitro* and *in vivo* [23], but produced hypolocomotion and ataxia in operant tasks[24-25]. However, chronic dosing of memantine significantly improved learning in mice without causing any psychomotor adverse effects, which implies that chronic dosing of memantine develops tolerance to sensorimotor side effects[26-27]. Therefore, the present study used chronic intraperitoneal (i.p.) administration of memantine in different doses to assess the efficacy of memantine on Aβ-mediated learning and memory disruption in rats. These findings provide further evidence of memantine as an AD treatment.

RESULTS

Quantitative analysis of experimental animals

A total of 42 rats were equally and randomly assigned to seven groups: control (i.p. injection of normal saline), 5, 20, 40 mg/kg memantine (i.p. injection of 5, 20, or 40 mg/kg memantine), Aβ treatment (i.p. injection of normal saline + hippocampal slices Aβ treatment), 5 or 20 mg/kg memantine + Aβ (i.p. injection of 5 or 20 mg/kg memantine + hippocampal slices Aβ treatment). The control and the 5, 20, and 40 mg/kg memantine groups were not treated with Aβ. All 42 rats were included in the final analysis.

Synthetic Aβ inhibited LTP induction in rat hippocampal slices

In our previous study, we found the threshold concentration of synthetic Aβ was 100–200 nM and a strong LTP inhibition was produced by 500 nM Aβ[6]. Therefore, we used a concentration of 500 nM synthetic $Aβ_{1-42}$ in the present study. In hippocampal slices of saline-treated rats, the average LTP induced by a brief high frequency stimulation (HFS) in the dentate gyrus measured 194 ± 9% and 148 ± 4% baseline at peak and 60 minutes post-HFS, respectively ($P < 0.01$, $n = 6$; Figure 1A). Perfusion of $Aβ_{1-42}$ (500 nM) for 40 minutes before HFS in hippocampal slices of saline-treated animals inhibited LTP, and the induction of LTP at peak

measured 152 ± 16% and measured 101 ± 3% at 60 minutes post-HFS baseline (both $P < 0.01$, $n = 6$; Figure 1B), which are significantly lower than those slices without Aβ treatment. These outcomes were consistent with our previous results that synthetic Aβ has an inhibitory effect on induction of hippocampal LTP.

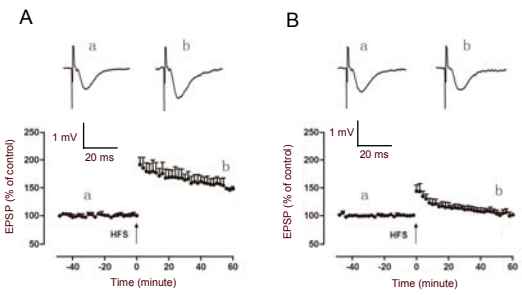

Figure 1 Synthetic amyloid-beta protein (Aβ) evokes potent inhibition of long-term potentiation (LTP) induction in rat hippocampal slices.

(A) Control LTP induced by a single brief high-frequency stimulation (HFS) in the dentate gyrus of rats intraperitoneally administered with normal saline ($n = 6$).

(B) LTP induction in the presence of synthetic Aβ (500 nM), bath-applied 40 minutes before HFS, were significantly reduced compared with controls ($n = 6$).

Data are expressed as mean ± SEM. The traces are field excitatory postsynaptic potentials (EPSPs) recorded before (a) and after (b) HFS.

Effects of chronic-treated memantine on LTP induction under physiological conditions

To examine whether chronic treatment of memantine affects normal learning function, we assessed the effects of chronic i.p. administration of various doses of memantine (5, 20, or 40 mg/kg per day for 7 days) on the induction of LTP in the rat hippocampus.

In memantine-treated (5 mg/kg per day) animals, the induction of LTP measured 193 ± 15% and 151 ± 8% baseline at peak and 60 minutes post-HFS, respectively, which was not significantly different from saline-treated controls ($P > 0.05$, $n = 6$; Figure 2A). Similarly, pretreatment with a dose of 20 mg/kg per day i.p. memantine, a typical therapeutic dose for AD treatment, did not affect LTP induction compared to saline controls (191 ± 12% and 146 ± 11% at peak and 60 minutes post-HFS, respectively; $P > 0.05$, $n = 6$; Figure 2B). However, animals treated with a higher dose (40 mg/kg per day, i.p.) of memantine showed complete abolishment of LTP induction, which measured 145 ± 3% and 108 ± 3% at peak and 60 minutes post-HFS ($P < 0.01$, $n = 6$; Figure 2C). The dose-response relationship of LTP induction and memantine concentration is shown in Figure 2D.

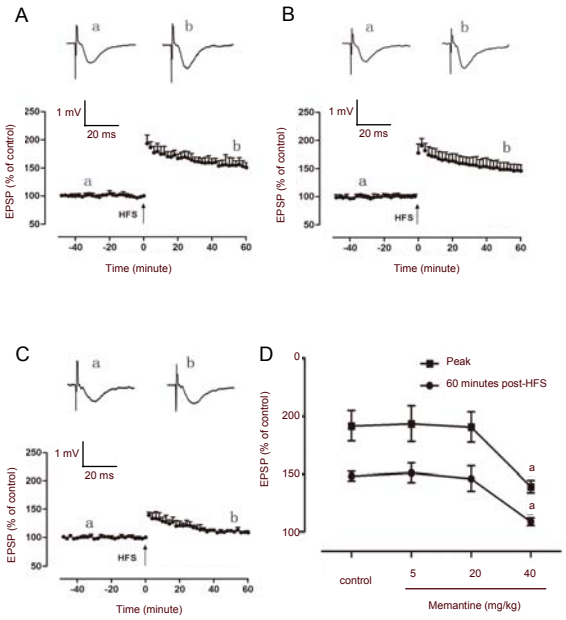

Figure 2 Concentration-effects of chronic memantine treatment on the induction of long-term potentiation (LTP).

(A) In the dentate gyrus of rats with repeated i.p. dosing of memantine (5 mg/kg for 7 days), high-frequency stimulation (HFS) induced LTP was not significantly different from control LTP.

(B) In the memantine-treated (20 mg/kg for 7 days, i.p.) rat hippocampus, LTP induction was not significantly different from control LTP.

(C) In memantine-injected (40 mg/kg for 7 days) animals, LTP induction was significantly reduced compared with controls.

(D) The potency curve of various i.p. doses of memantine on the induction of LTP at both peak and 60 minutes post-HFS. Data are expressed as mean ± SEM of six rats in each group. The traces are field excitatory postsynaptic potentials (EPSPs) recorded before (a) and after (b) HFS. [a]$P < 0.01$, *vs*. control group (analysis of variance).

Protective effects of chronic-treated memantine on Aβ-mediated LTP inhibition *in vitro*

Because the induction of hippocampal LTP was greatly inhibited in 40 mg/kg memantine-treated (i.p for 7 days) rats (Figure 2C), it is possible that chronic treatment of 40 mg/kg memantine may cause memory defects in physiological states. Therefore, we only tested the effects of lower doses of memantine (5 and 20 mg/kg) on Aβ-induced LTP inhibition. Chronic treatment of animals with low dose of memantine (5 mg/kg) did not affect Aβ-mediated inhibition of LTP which measured 144 ± 11% and 106 ± 5% baseline at peak and 60 minutes post-HFS, respectively, which was not significantly different from the Aβ-treated control ($P > 0.05$, $n = 6$; Figure 3A). However, chronic treatment of animals with

20 mg/kg memantine (i.p. for 7 days) strongly prevented the inhibitory effect of $A\beta_{1-42}$ on LTP induction. LTP measured 187 ± 18% and 129 ± 8% baseline at peak and 60 minutes post-HFS, respectively (Figure 3B) when compared with the Aβ-treated control ($P < 0.01$, $n = 6$; a sample of control shown as Figure 1B), although the values at 60 minutes post-HFS were significantly lower than saline-treated control values ($P < 0.05$, $n = 6$).

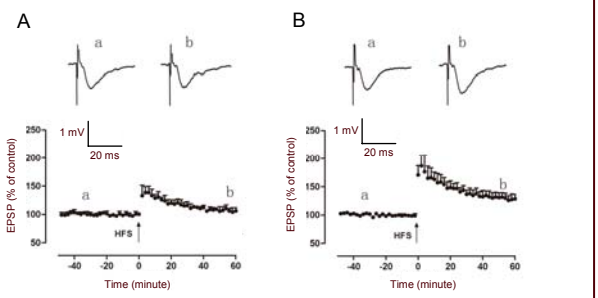

Figure 3 Effects of chronic treatment with memantine on amyloid-beta protein (Aβ)-triggered long-term potentiation (LTP) impairment in rat hippocampal slices.

(A) In animals pretreated with intraperitoneal injection of memantine (5 mg/kg for 7 days, $n = 6$), the incubation of $A\beta_{1-42}$ (500 nM) completely inhibited the induction of LTP.

(B) In rats treated with intraperitoneal injection of memantine (20 mg/kg for 7 days, $n = 6$), the perfusion of $A\beta_{1-42}$ (500 nM) only partially inhibited LTP.

All values were expressed as mean ± SEM. The traces are field excitatory postsynaptic potentials (EPSPs) recorded before (a) and following (b) high-frequency stimulation.

DISCUSSION

Memantine is a voltage-dependent NMDA receptor antagonist and is used as a treatment of moderate-to-severe AD. Clinical trials have demonstrated cognitive and behavioral improvements after a few weeks of memantine treatment[28-29]. Results from animal studies clearly show that memantine has a broad range of effects. It reverses the recognition memory deficits in aged rats[30] and enhances spatial memory in healthy animals[27]. While some studies reported improved spatial cognition by memantine, others reported memantine-induced cognitive deficits or no effect on spatial memory[25, 31]. In the present study, we demonstrated that chronic treatment of memantine at a dose within therapeutic range (20 mg/kg, i.p. for 7 days) reversed the Aβ-induced inhibitory effect on hippocampal LTP in rat brain without interrupting LTP induction under physiological conditions. A lower dose (5 mg/kg) did not have a protective role against the toxic effect of Aβ on LTP induction while a higher dose of memantine (40 mg/kg) inhibited LTP in control rats. These outcomes suggest that long-term systemic administration of memantine within a certain dose range could oppose Aβ-related dementia in affected brain regions, yet may not interrupt normal memory and cognitive tasks in functional brain regions.

Excessive or inappropriate activation of NMDARs can disrupt synaptic plasticity[32]. Memantine is reported to bind to human cortical NMDARs with a *Ki* of approximately 0.5 μM and inhibit NMDARs with an IC_{50} of approximately 1 μM[27]. In clinical practice, a stable dose of memantine (20 mg per day) has been found to produce a steady-state plasma drug level of approximately 0.5 μM in AD patients[33]. Considerable evidence indicates that memantine plays a potent neuroprotective role in different models of neurotoxicity *in vitro* [15-19]. However, other studies have shown that memantine, when applied acutely or semi-chronically, has a disruptive effect on memory tasks in different animal models[20-23], and this disruptive role may result from memantine blockage of NMDARs. Acute i.p. doses of 2.5–5 mg/kg memantine resulted in approximately 1 μM plasma memantine concentration and has been found to improve cognitive function in mice[34]. However, moderate hypolocomotion and/or ataxia have been observed in rodents after acute i.p. injection of memantine at doses below 30 mg/kg[24-25], suggesting that even acute moderate doses of memantine may disrupt the normal functioning of NMDARs and alter learning in healthy individuals[35]. Different doses of memantine may have various effects. Chronic treatment of memantine (10 and 30 mg/kg for 4 weeks) improved hippocampus-related spatial learning in a transgenic mouse model of AD without notable locomotion/exploratory defects[34]. Similar results were also reported in AD mice after long-term administration of memantine (30 mg/kg daily for 5 weeks)[36]. Interestingly, oral dosing of memantine (20 mg/kg per day for 8 days) significantly reduced the cortical levels of soluble $A\beta_{1-42}$ in APP/PS1 transgenic mice[16], while a higher dose of memantine (100 mg/kg daily for 4 weeks) improved cognition in mice without psychomotor side effects[27]. Consistent with previous outcomes, the present study demonstrated that chronic memantine application at a 20 mg/kg dose may avoid memory disruption because 7-day i.p. application at this dose did not inhibit hippocampal LTP.

Memantine may function by antagonizing extrasynaptic NMDARs[37]. Extrasynaptic NMDARs have been reported to preferentially mediate the toxic effects of excessive glutamate and to facilitate the aggregation of misfolded proteins to form a more toxic pattern[38]. Memantine, at

therapeutic concentrations, improved the balance of excitatory activity by preferentially blocking extrasynaptic NMDARs, even under pathologically depolarizing conditions, while relatively sparing synaptic communication[39]. Memantine may exert its neuroprotective effects by lowering the extracellular Mg^{2+} concentration. Memantine may also be involved in NMDAR independent mechanisms. Memantine depressed evoked glutamate release *via* presynaptic voltage-dependent Ca^{2+} channels and subsequently suppressed the protein kinase C signaling cascade. Interestingly, these results do not relate with the contribution of NMDARs in rat cerebrocortical nerve terminals[40]. Also, Ca^{2+}-dependent protein kinase C signaling may be involved in metabotropic glutamate receptor-dependent LTP. Such metabotropic glutamate receptor-related LTP can be expressed presynaptically or postsynaptically, but may involve co-activation of other receptors, such as NMDAR, dopamine or adenosine receptors[41]. Further exploration is required to determine whether the beneficial effects of memantine against $A\beta_{1-42}$-mediated LTP inhibition are associated with NMDARs or other receptors.

In summary, chronic treatment with memantine in a certain dose range can attenuate the $A\beta_{1-42}$-induced rapid disruption of hippocampal LTP *in vitro*. In addition, high dose of i.p. administered memantine potently inhibited induction of LTP in rats. These findings may provide a better understanding of memantine in AD treatment and provide more information on effective disease-modifying therapeutics.

MATERIALS AND METHODS

Design

A randomized, controlled animal study and *in vitro* electrophysiological observation.

Time and setting

The experiment was performed at Ningbo University, China and Trinity College Dublin, Ireland, between 2009 and 2010.

Materials

A total of 42 male Wistar rats, weighing 40–80 g and aged 3–4 weeks, were obtained from the Animal House of Trinity College, Ireland and Shanghai Silaike Experimental Animal Co., Ltd. (certificate No. SCXK (Hu) 2007-0005). The use of animals for experimental procedures was conducted in accordance with the *Guidance Suggestions for the Care and Use of Laboratory Animals*, issued by the Ministry of Science and Technology of China[42].

Methods

Drug treatment

Memantine hydrochloride was purchased from Sigma (St. Louis, MO, USA). For the experiments, memantine (5, 20, and 40 mg/kg) was prepared for i.p. administration in normal saline (0.9%). Memantine was administered i.p. for 7 days at doses of 5, 20 or 40 mg/kg per day. The control group received the same volume of i.p. dosing of normal saline.

Preparation of hippocampal slices

Transverse slices of the hippocampus were prepared from memantine- or saline-treated rats. The brains were rapidly removed after the rats were sacrificed and placed in cold oxygenated (95% O_2/5% CO_2) physiological media. Slices were cut at a thickness of 350 μM using an Intracell Plus 1000 vibratome (Vibratome Co., St. Louis, MO, USA) and placed in a storage container containing oxygenated medium (self made) at 20–22°C for 1 hour. The slices were then transferred to a recording chamber for submerged slices and continuously superfused at a rate of 5–6 mL/min at 30–32°C. The control media contained NaCl 120 mM; KCl 2.5 mM, NaH_2PO_4 1.25 mM; $NaHCO_3$ 26 mM; $MgSO_4$ 2.0 mM; $CaCl_2$ 2.0 mM; and D-glucose 10 mM. All solutions contained 100 μM picrotoxin (Sigma) to block $GABA_A$-mediated activity.

Aβ treatment

In experiments involving the application of $A\beta_{1-42}$, Aβ was perfused for 40 minutes before HFS. Synthetic $A\beta_{1-42}$ (Bachem, Bubendorf, Switzerland) was prepared as a stock solution of 50 μM in ammonium hydroxide (0.1%), stored at –20°C, and then added to the physiological medium immediately prior to each experiment. Control (0.9% saline-injected) and experimental levels of LTP were measured on slices prepared from the same hippocampus.

In vitro electrophysiological recording

Standard electrophysiological techniques were used to record field potentials[6]. Presynaptic stimulation was applied to the medial perforant pathway of the dentate gyrus using a bipolar insulated tungsten wire electrode (Sutter Instrument Company, Novato, CA, USA), and field excitatory postsynaptic potentials (EPSPs) were recorded at a control test frequency of 0.033 Hz from the middle one-third of the molecular layer of the dentate gyrus with a glass microelectrode (Sutter Instrument

Company). The inner blade of the dentate gyrus was used in all studies. In each experiment, an input-output curve (afferent stimulus intensity versus EPSP amplitude) was plotted at the test frequency. For all experiments, the amplitude of the test EPSP was adjusted to one-third of maximum (–1.2 mV). LTP was evoked by HFS consisting of eight trains, each consisting of eight stimuli at 200 Hz, with inter-train intervals of 2 seconds. The stimulation voltage was increased during the HFS to evoke an initial EPSP of the train of double the normal test EPSP amplitude. Control (0.9% saline-treated) and experimental levels of LTP were measured on slices prepared from the same hippocampus.

Statistical analysis

Recordings were analyzed using p-CLAMP (Axon Instruments, Sunnyvale, CA, USA). Data were expressed as mean ± SEM. Analysis of variance was used for statistical comparison. $P < 0.05$ was considered statistically significant.

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Author contributions: Qinwen Wang designed and revised the manuscript. Fushun Li performed the experiments. Xiaowei Chen conducted the experiments and wrote the manuscript. Feiming Wang provided computer science support. Shujun Xu and Lan Chang provided technological support. Roger Anwyl supervised the study. All authors approved the final version of the paper.

Conflicts of interest: None declared.

Ethical approval: The study was approved by the Guidelines for the Care and Use of Laboratory Animals of Ningbo University, China.

Author statements: The manuscript is original, has not been submitted to or is not under consideration by another publication, has not been previously published in any language or any form, including electronic, and contains no disclosure of confidential information or authorship/patent application/funding source disputations.

REFERENCES

[1] Hardy J. A hundred years of Alzheimer's disease research. Neuron. 2006;52(1):3-13.

[2] Walsh DM, Klyubin I, Fadeeva JV, et al. Naturally secreted oligomers of amyloid beta protein potently inhibit hippocampal long-term potentiation in vivo. Nature. 2002;416(6880):535-539.

[3] Selkoe DJ. Soluble oligomers of the Amyloid β-protein impair synaptic plasticity and behavior. Behav Brain Res. 2008;192(1):106-113.

[4] Small DH, Mok SS, Bornstein JC. Alzheimer's disease and Abeta toxicity: from top to bottom. Nat Rev Neurosci. 2001;2(8):595-598.

[5] Cleary JP, Walsh DM, Hofmeister JJ, et al. Natural oligomers of the amyloid-beta protein specifically disrupt cognitive function. Nat Neurosci. 2005;8(1):79-84.

[6] Wang Q, Walsh DM, Rowan MJ, et al. Block of long-term potentiation by naturally secreted and synthetic amyloid beta-peptide in hippocampal slices is mediated via activation of c-Jun N-terminal kinase, cyclin-dependent kinase 5, and p38 mitogen-activated protein kinase as well as metabotropic glutamate receptor type 5. J Neurosci. 2004;24(13):3370-3378.

[7] Hock C, Konietzko U, Streffer JR, et al. Antibodies against beta-amyloid slow cognitive decline in Alzheimer's disease. Neuron. 2003;38(4):547-554.

[8] Klyubin I, Walsh DM, Lemere CA, et al. Amyloid beta protein immunotherapy neutralizes Abeta oligomers that disrupt synaptic plasticity in vivo. Nat Med. 2005;11(5):556-561.

[9] Schenk DB, Seubert P, Grundman M, et al. Abeta immunotherapy: lessons learned for potential treatment of Alzheimer's disease. Neurodegener Dis. 2005;2(5):255-260.

[10] Lipton SA. Failures and successes of NMDA receptor antagonists: molecular basis for the use of open-channel blockers like Memantine in the treatment of acute and chronic neurologic insults. NeuroRx. 2004;1(1):101-110.

[11] Seow D, Gauthier S. Pharmacotherapy of Alzheimer disease. Can J Psychiatry. 2007;52(10):620-629.

[12] Lipton SA. Pathologically activated therapeutics for neuroprotection. Nat Rev Neurosci. 2007;8(10):803-808.

[13] Parsons CG, Stöffler A, Danysz W. Memantine: a NMDA receptor antagonist that improves memory by restoration of homeostasis in the glutamatergic system-too little activation is bad, too much is even worse. Neuropharmacology. 2007;53(6):699-723.

[14] Raina P, Santaguida P, Ismaila A, et al. Effectiveness of cholinesterase inhibitors and memantine for treating dementia: evidence review for a clinical practice guideline. Ann Intern Med. 2008;148(5):379-397.

[15] Miguel-Hidalgo JJ, Alvarez XA, Cacabelos R, et al. Neuroprotection by memantine against neurodegeneration induced by beta-amyloid(1-40). Brain Res. 2002;958(1):210-221.

[16] Alley GM, Bailey JA, Chen D, et al. Memantine lowers amyloid-beta peptide levels in neuronal cultures and in APP/PS1 transgenic mice. J Neurosci Res. 2010;88(1):143-154.

[17] Chipana C, Torres I, Camarasa J, et al. Memantine protects against amphetamine derivatives-induced neurotoxic damage in rodents. Neuropharmacology. 2008;54(8):1254-1263.

[18] Babu CS, Ramanathan M. Pre-ischemic treatment with memantine reversed the neurochemical and behavioural parameters but not energy metabolites in middle cerebral artery occluded rats. Pharmacol Biochem Behav. 2009; 92(3):424-432.
[19] Hare WA, Wheeler L. Experimental glutamatergic excitotoxicity in rabbit retinal ganglion cells: block by memantine. Invest Ophthalmol Vis Sci. 2009;50(6): 2940-2948.
[20] Barber TA, Haggarty MK. Memantine ameliorates scopolamine-induced amnesia in chicks trained on taste-avoidance learning. Neurobiol Learn Mem. 2010;93(4):540-545.
[21] Barnes CA, Danysz W, Parsons CG.. Effects of the uncompetitive NMDA receptor antagonist memantine on hippocampal long-term potentiation, short-term exploratory modulation and spatial memory in awake, freely moving rats. Eur J Neurosci. 1996;8(3):565-571.
[22] Zajaczkowski W, Frankiewicz T, Parsons CG, et al. Uncompetitive NMDA receptor antagonists attenuate NMDA-induced impairment of passive avoidance learning and LTP. Neuropharmacology. 1997;36(7):961-971.
[23] Klyubin I, Wang Q, Reed MN, et al. Protection against Aβ-mediated apid disruption of synaptic plasticity and memory by memantine. Neurobiol Aging. 2009;32(4): 614-623.
[24] Kos T, Popik P. A comparison of the predictive therapeutic and undesired side-effects of the NMDA receptor antagonist, memantine, in mice. Behav Pharmacol. 2005; 16(3):155-161.
[25] Creeley C, Wozniak DF, Labruyere J, et al. Low doses of memantine disrupt memory in adult rats. J Neurosci. 2006; 26(15):3923-2932.
[26] Minkeviciene R, Banerjee P, Tanila H. Memantine improves spatial learning in a transgenic mouse model of Alzheimer's disease. J Pharmacol Exp Ther. 2004;311(2): 677-682.
[27] Minkeviciene R, Banerjee P, Tanila H. Cognition-enhancing and anxiolytic effects of memantine. Neuropharmacology. 2008;54(7):1079-1085.
[28] Peskind ER, Potkin SG, Pomara N, et al. Memantine treatment in mild to moderate Alzheimer disease: a 24-week randomized, controlled trial. Am J Geriatr Psychiatry. 2006;14(8):704-715.
[29] Schulz JB, Rainer M, Klünemann HH, et al. Sustained effects of once-daily memantine treatment on cognition and functional communication skills in patients with moderate to severe Alzheimer's disease: results of a 16-week open-label trial. J Alzheimers Dis. 2011;25(3): 463-475.
[30] Pietá Dias C, Martins de Lima MN, Presti-Torres J, et al. Memantine reduces oxidative damage and enhances long-term recognition memory in aged rats. Neuroscience. 2007;146(4):1719-1725.
[31] Quan MN, Zhang N, Wang YY, et al. Possible antidepressant effects and mechanisms of memantine in behaviors and synaptic plasticity of a depression rat model. Neuroscience. 2011;182:88-97.
[32] Zorumski CF, Izumi Y. Modulation of LTP induction by NMDA receptor activation and nitric oxide release. Prog Brain Res. 1998;118:173-182.
[33] Witt A, Macdonald N, Kirkpatrick P. Memantine hydrochloride. Nat Rev Drug Discov. 2004;3(2):109-110.
[34] Zoladz PR, Campbell AM, Park CR, et al. Enhancement of long-term spatial memory in adult rats by the noncompetitive NMDA-receptor antagonists, memantine and neramexane. Pharmacol Biochem Behav. 2006;85(2): 298-306.
[35] Lockrow J, Boger H, Bimonte-Nelson H, et al. Effects of long-term memantine on memory and neuropathology in Ts65Dn mice, a model for Down syndrome. Behav Brain Res. 2011;221(2):610-622.
[36] Arif M, Chikuma T, Ahmed MM, et al. Effects of memantine on soluble Aβ25-35-induced changes in peptidergic and glial cells in Alzheimer's disease model rat brain regions. Neuroscience. 2009;164(3):1199-1209.
[37] Kotermanski SE, Johnson JW. Mg^{2+} imparts NMDA receptor subtype selectivity to the Alzheimer's drug memantine. J Neurosci. 2009;29(9):2774-2779.
[38] Okamoto S, Pouladi MA, Talantova M, et al. Balance between synaptic versus extrasynaptic NMDA receptor activity influences inclusions and neurotoxicity of mutant huntingtin. Nat Med. 2009;15(12):1407-1413.
[39] Xia P, Chen HS, Zhang D, et al. Memantine Preferentially Blocks Extrasynaptic over Synaptic NMDA Receptor Currents in Hippocampal Autapses. J Neurosci. 2010; 30(33):11246-11250.
[40] Lu CW, Lin TY, Wang SJ. Memantine depresses glutamate release through inhibition of voltage-dependent Ca2+ entry and protein kinase C in rat cerebral cortex nerve terminals: An NMDA receptor-independent mechanism. Neurochem Int. 2010;57(2):168-176.
[41] Anwyl R. Metabotropic glutamate receptor-dependent long-term potentiation. Neuropharmacology. 2009;56(4): 735-740.
[42] The Ministry of Science and Technology of the People's Republic of China. Guidance Suggestions for the Care and Use of Laboratory Animals. 2006-09-30.

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