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Acute biphasic dose-response effects of amantadine hydrochloride on bladder storage in conscious rats

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Abstract

Parkinson's disease (PD) causes prevalent lower urinary tract (LUT) dysfunction such as overactive bladder and severely impairs patients' quality of life. Current management of LUT dysfunction in PD remains unsatisfactory. Amantadine hydrochloride, an anti-Parkinson drug, improves PD movement symptoms clinically, yet its urodynamic dose-response is unclear. This study adopted continuous cystometry in normal male Sprague-Dawley rats to explore amantadine's bladder regulatory effects. Forty rats were assigned to saline control, 0.3, 3, 30, 100 μmol/kg groups. Four core urodynamic indicators—maximum void volume (MV), inter-void interval (IT), maximum detrusor contraction pressure (MP) and post-void residual volume (RV)—were calculated as fold change against individual baseline. 3 μmol/kg amantadine significantly raised MV and prolonged IT, improving bladder storage capacity. Conversely, 100 μmol/kg extreme dose markedly reduced MV and shortened IT. No dose induced statistically significant changes in MP or RV, indicating amantadine solely regulates bladder storage without impairing voiding efficiency. This work first confirms amantadine exerts non-monotonic biphasic modulation on rodent LUT storage function in vivo. Clinicians may optimize oral amantadine dosage to relieve PD storage LUT dysfunction.

Introduction

PD is defined by motor symptoms including bradykinesia, rigidity and resting tremor, yet non-motor LUT dysfunction also strongly reduces patients' quality of life. Epidemiological data show 50–90% PD patients develop storage-phase LUT disorders (frequency, urgency, low void volume) consistent with OAB [1.2].

Current OAB therapies for PD mainly use peripheral β3 agonists like mirabegron, failing to act on pontine micturition regulatory centers. Levodopa and apomorphine ease storage LUT symptoms in PD patients, highlighting the therapeutic value of central dopaminergic modulation of micturition pathways [3], but severe emesis and hypotension limit its clinical use. Blood-brain barrier-permeable anti-Parkinson agents may exert unrecognized bladder regulatory effects.

Amantadine hydrochloride, originally an antiviral agent approved in 1968 and later a first- or second-line PD therapy [4.5], alleviates clinical PD movement symptoms, but no controlled animal studies have clarified its urodynamic characteristics. This study applied continuous awake cystometry in unrestrained rats to fully characterize amantadine's dose-related impacts on core bladder parameters.

Materials and Methods

Animal Model Selection and Group Allocation

Forty 8–9 week-old male Sprague-Dawley rats (250–300 g) were randomly divided into five groups with further intravenous amantadine doses (0.3, 3, 30, 100 μmol/kg) or saline control. All operations

minimized animal suffering; the protocol obtained institutional ethical approval and fully complied with ARRIVE guidelines.

Surgical Catheter Implantation

Three days before testing, rats received intraperitoneal pentobarbital anesthesia (40 mg/kg). PE-50 catheters were implanted into the bladder dome for intravesical pressure recording and jugular vein for drug injection, then tunnelled subcutaneously and fixed on the scalp. Daily intravenous penicillin (0.05 mL/kg) was administered for 3 days postoperatively until incision healing. Rats were single-housed under a 12 h light-dark cycle with free food and water for acclimatization.

Awake Urodynamic Recording Protocol

Urodynamic detection started at 20:00 without anesthesia or restraint. Intravesical catheters were connected to a pressure transducer and infusion pump; saline perfused at 12 mL/h to mimic physiological urine production. An electronic balance continuously recorded voided volume in real time. Each rat underwent 60 min baseline recording before drug injection, followed by another 60 min post-dose recording. maximum void volume (MV) and inter-void interval (IT) reflected storage capacity; maximum detrusor contraction pressure (MP) and post-void residual volume (RV) represented voiding efficiency. All post-dose data were normalized to individual baseline (fold changes set to 1.0 for baseline).

Statistical Analysis

Intra-animal baseline vs post-dose comparisons were performed with Excel Statistics Excel Statistics version 6.0. Data were expressed as mean ± SD. Repeated-measures ANOVA and paired t-test were adopted for intergroup and intra-animal analysis, with $P < 0.05$ set as statistical significance threshold.

At the end of all experimental recordings, all rats were humanely euthanized via intraperitoneal injection of overdose sodium pentobarbital (≥ 100 mg/kg). Complete death was confirmed by permanent cessation of respiration and absence of corneal reflex.

Results

Effects on Bladder Storage Function (MV and IT)

Compared to baseline, the saline control group showed no significant change in MV (1.01 ± 0.09, $P > 0.05$). The low dose (0.3 μmol/kg) produced a slight increase that did not reach statistical significance (1.07 ± 0.07, $P > 0.05$). The medium dose (3 μmol/kg) significantly increased MV (1.23 ± 0.07, $P < 0.05$). The high dose (30 μmol/kg) showed no significant effect (1.07 ± 0.11, $P > 0.05$). In contrast, the extreme dose (100 μmol/kg) significantly decreased MV (0.67 ± 0.08, $P < 0.05$) (Fig. 1).

A similar pattern was observed for IT. The saline control group showed no significant change (0.99 ± 0.13, P > 0.05). The low dose showed a slight increase without statistical significance (1.06 ± 0.04, P > 0.05). The medium dose significantly prolonged IT (1.28 ± 0.13, P < 0.05). The high dose produced no significant change (0.95 ± 0.10, P > 0.05). The extreme dose showed a marked decrease that did reach statistical significance (0.54 ± 0.09, P < 0.05) (Fig. 2).

These results demonstrate a biphasic dose–response relationship: moderate amantadine enhances storage function, while a suprapharmacological dose impairs it.

Effects on Voiding Function (MP and RV)

No significant changes in MP were observed at any dose. The saline control group (0.92 ± 0.13), low dose (0.94 ± 0.11), medium dose (0.91 ± 0.27), high dose (0.90 ± 0.36), and extreme dose (0.79 ± 0.31) all showed no statistically significant difference from baseline (P > 0.05 for all).

Similarly, no significant changes in post-void residual volume were observed at any dose. The saline control group (1.00 ± 0.03), low dose (1.00 ± 0.05), medium dose (1.01 ± 0.04), high dose (1.02 ± 0.06), and extreme dose (1.22 ± 0.63) all remained close to baseline, with no statistical significance (P > 0.05 for all).

Data curves of MP and RV are attached as supplementary figures (Supplementary Figure S1, S2).

Therefore, amantadine at any tested dose did not alter detrusor contractility or voiding efficiency, its effects were confined to the storage phase.

Discussion

This conscious rat urodynamic research first proves amantadine exerts acute dose-dependent biphasic regulation on bladder storage function in rats. Moderate 3 μmol/kg dose improves storage capacity via larger MV and longer IT; extreme dose 100 μmol/kg amantadine impairs storage via reduced MV and IT. This dose-response curve provides pharmacological evidence explaining amantadine’s clinical bladder benefits.

The biphasic bladder responses in healthy rats interpret Sakakibara’s clinical observations that oral amantadine reduces micturition frequency and relieves storage LUT symptoms in PD patients. Clinical symptom improvement in his study precisely matches our medium-dose experimental outcomes, forming complementary evidence[6].

Amantadine’s biphasic profile resembles apomorphine, a non-selective dopamine agonist that elevates bladder capacity at low doses and suppresses storage at high doses [3]. However, apomorphine’s clinical application is restricted by severe vomiting, hypotension and elevated residual urine volume—side effects absent in our amantadine datasets. Notably, all tested amantadine doses fail to alter MP and RV

significantly, confirming its pharmacological activity exclusively targets storage pathways without impairing voiding efficiency. That distinction matters clinically because those long-standing PD patients may not only storage problems but also have detrusor hypocontractility.

Amantadine's dual anti-Parkinson mechanism combining dopaminergic potentiation and NMDA receptor antagonism jointly mediates its unique bladder regulatory effects [4, 7]. Within micturition circuits, D1 receptors suppress voiding reflexes while D2 receptors trigger detrusor overactivity; this D1/D2 balance model accounts for apomorphine's biphasic effects [8–10]. Nevertheless, our data from neurologically intact rats indicate central dopaminergic signalling cannot fully explain amantadine's bladder regulation.

As a weak NMDA antagonist (Ki = 10 μM), amantadine modulates spinal afferent micturition pathways to adjust voiding reflex sensitivity. [2]. Meanwhile, clinical anticholinergic adverse events like urinary retention imply amantadine acts directly on peripheral bladder smooth muscle, a hypothesis requiring isolated tissue validation [11] .Its pleiotropic glutamatergic and dopaminergic activities together drive biphasic bladder responses, which cannot be simplified to D1/D2 receptor balance alone.

Unlike levodopa and apomorphine, amantadine maintains normal contraction pressure and residual urine across all doses, separating storage regulation from voiding dysfunction. Long-term high-dose levodopa elevates bladder outlet resistance and residual volume in PD patients, mainly due to excessive D2-mediated detrusor overactivity [12, 13, 14]. Though D2 activation relaxes urethral sphincters to partially counteract detrusor contraction [23], this compensation fails in most levodopa-treated patients. We infer amantadine's combined dopaminergic enhancement and NMDA inhibition generates balanced bladder-urethra regulatory output, avoiding voiding defects induced by single dopamine agonist monotherapy.

Clinical Implications and Experimental Limitations

Amantadine is widely approved for PD motor symptoms, enabling clinicians to optimize its oral dosage to treat PD-related storage LUT dysfunction without novel drug development, offering a low-cost, rapid therapeutic strategy applicable to non-PD OAB disorders. Four core limitations restrict direct clinical translation: (i) experiments used healthy rats rather than PD lesion models; (ii) rodent and human pharmacokinetics differ greatly, preventing direct dose extrapolation; (iii) respective contributions of dopaminergic and glutamatergic pathways remain unquantified; (iv) only acute single-dose effects were detected, lacking chronic long-term observation. Future research priorities include cystometry in 6-OHDA PD rats, clinical dose-escalation trials with urodynamic monitoring, and selective receptor blocking tests to quantify pathway contributions, so as to define safe and effective clinical dosage windows for amantadine in LUT dysfunction management.

Declarations

Funding and/or Conflicts of interests/Competing interests

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Conflicts of interest/Competing interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Ethics approval

All animal experiments were approved by the Ethics Committee in Beihua Universit (Approval No. 2026081115). All animal experimental procedures were approved by the Institutional Animal Care and Use Committee and were performed in strict accordance with the ARRIVE guidelines and national regulations for the care and use of laboratory animals.

Consent for publication

Not applicable.

Availability of data and material

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Authors' contributions

[Yang Liu]: Conceptualization, Methodology, Investigation, Writing – original draft,Funding acquisition.

[Yanlong Mu]: Methodology, Data curation, Formal analysis.

[Haoyun Liu]: Methodology, Data curation, Formal analysis.

[Jiahui Jin]: Methodology, Data curation, Formal analysis.

[Ke Sun]: Methodology, Data curation, Formal analysis.

[Jun Ren]: Methodology, Data curation, Formal analysis.

[Zhi Liu]: Conceptualization, Writing – review & editing,

Supervision.

All authors read and approved the final manuscript.

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Figures

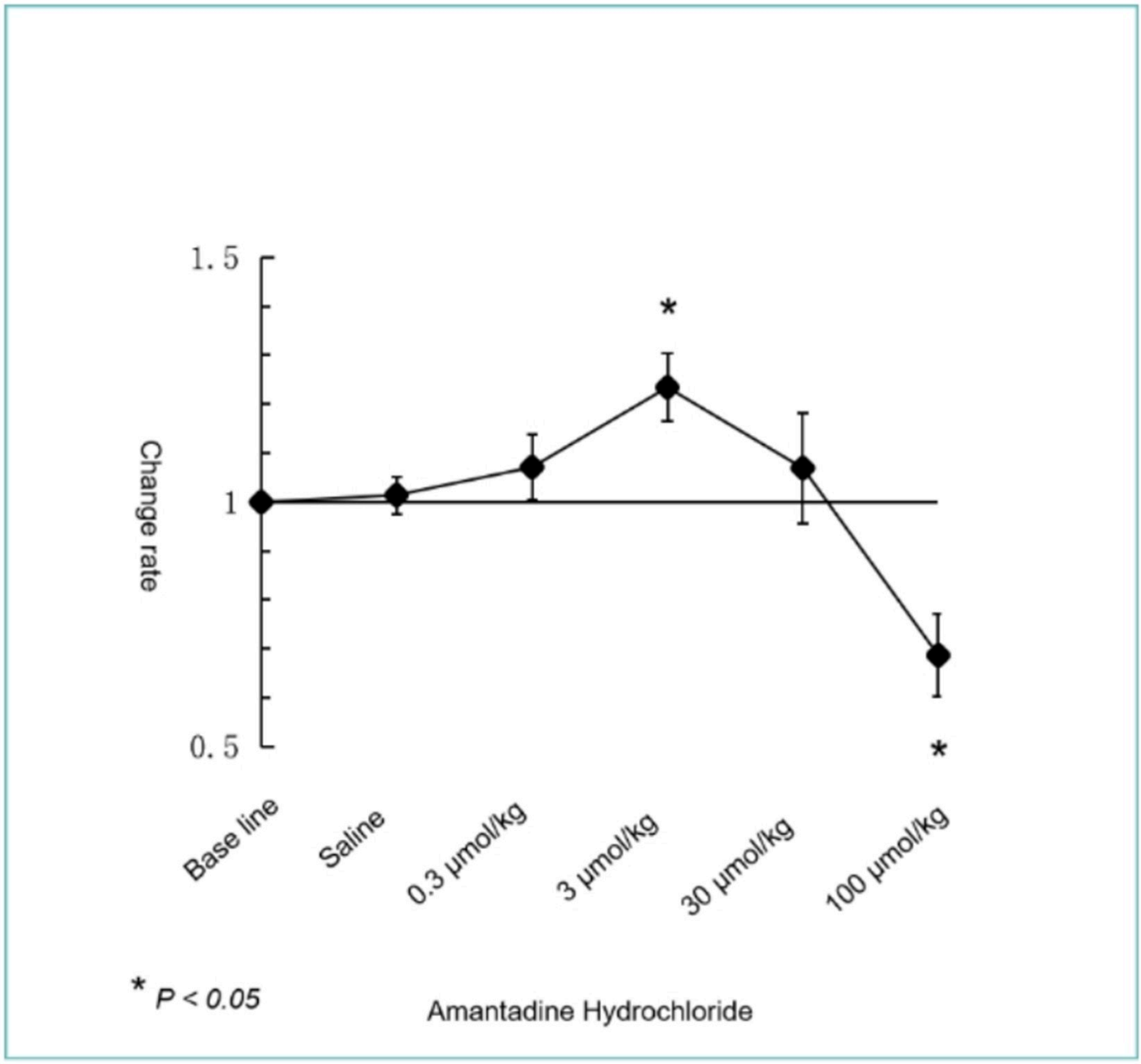


Figure 1

Effects of single-dose amantadine hydrochloride on MV

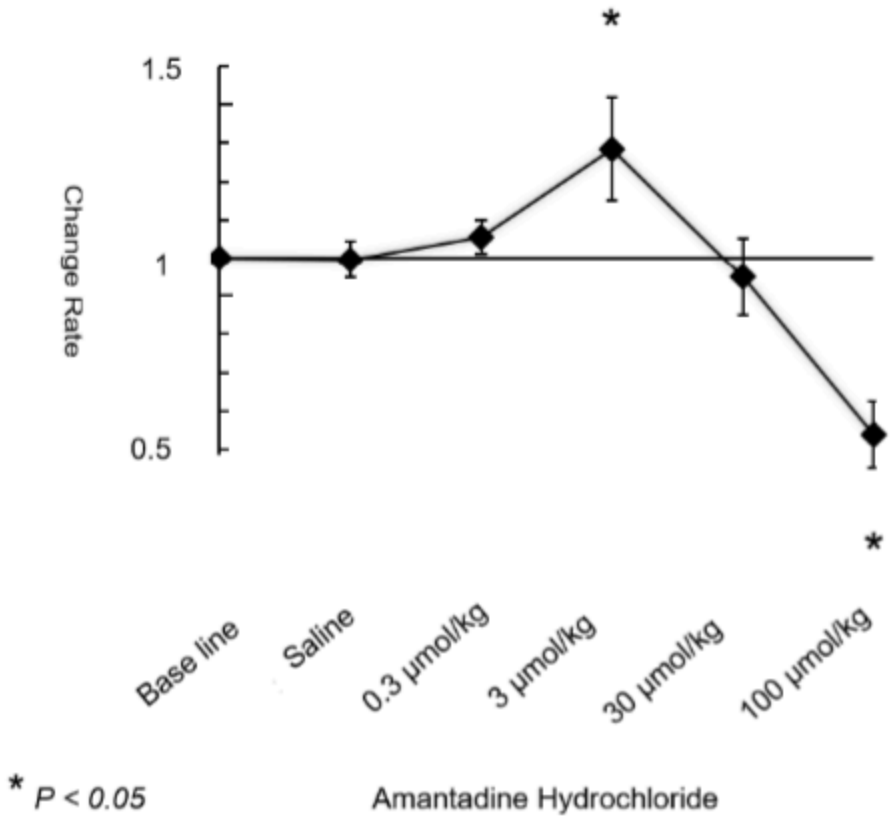


Figure 2

Effects of single-dose amantadine hydrochloride on IT

Supplementary Files

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- SupplementaryFigure.docx