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EXPERIMENTAL PAPER

Berberis vulgaris alleviates Levodopa-induced dyskinesia in male mice

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Summary

Introduction: Parkinson's disease is a chronic debilitating disease and many patients use Levodopa as a major treatment. However, this drug in long-term use causes a serious condition that is known as Levodopa-induced dyskinesia (LID). *Berberis vulgaris* (BV) has been known to be a good potential medication for neurologic diseases such as movement disorders. The aim of this study is to investigate the usefulness of BV for LID in mice.

Material and methods: In this study, 48 adult male mice were randomly divided into six groups: 1) saline group, 2) MPTP + LID, 3) MPTP + LID + BV (5 mg/kg), 4) MPTP + LID + BV (10 mg/kg), 5) MPTP + LID + BV (20 mg/kg). MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) (30 mg/kg/ day/*i.p.*) was used to induce Parkinson's disease and Levodopa (50 mg/kg/day/*i.p.*) was used to induce LID. After induction of LID, mice received intraperitoneally (*i.p.*) different dosages of BV for 25 days. To investigate movement disorder improvement (dyskinesia), AIMS (Abnormal Involuntary Movement Scale) and cylinder tests were used.

Results: Mice that received BV at dosages of 10 and 20 mg/kg/day showed improvement in AIMS and the cylinder test.

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Conclusion: BV is a useful drug for treating LID. So, parkinsonian disease patients may get a beneficial effect after treatment with BV for LID.

Key words: **Parkinson's disease, Levodopa induced dyskinesia, *Berberis vulgaris*, dyskinesia, cylinder test, AIMS test**

Słowa kluczowa: **choroba Parkinsona, dyskineza indukowana L-dopą, *Berberis vulgaris*, dyskineza, test cylindra, test AIMS**

INTRODUCTION

Parkinson's disease is a neurodegenerative and debilitating disease that mostly affects the motor system. Motor symptoms are shaking, rigidity, slowness of movement and difficulty in walking [1]. As motor symptoms mostly occur during earlier phases of the disease, drug therapy is also targeted at curing motor symptoms [2]. Other symptoms appear in later stages of the disease. The most effective therapy for motor symptoms is application of a dopamine agonist such as L-dopa [3]. At earlier stages of Parkinson's disease, L-dopa is an effective therapy for alleviation of motor symptoms, but, with long term treatment, L-dopa-induced dyskinesia develops [4].

Medicinal plants recently have got much interest in treating various diseases [5]. *Berberis vulgaris* (BV) is a kind of fruit that is produced in many different countries [6]. The effectiveness of this plant has been largely investigated in recent decades. The effectiveness of this plant in treating disease is related to its alkaloid compound that has antioxidant and anti-inflammatory properties [5]. In this study, *B. vulgaris* has been investigated at various dosages for its usefulness for curing L-dopa-induced dyskinesia in animal model of Parkinson's disease.

The aim of this study was to investigate whether various doses of fruit water extract of *B. vulgaris* are effective in reducing L-dopa-induced dyskinesia.

MATERIAL AND METHODS

Animal care: Male mice (20–25 g) were kept in controlled conditions (12 h light/12 h dark cycle at a controlled temperature (22±2°C) and humidity (50±5%)) in plexiglass cages provided with *ad libitum* access to food and water. Animals were handled, trained and tested during the light cycle. All experimental protocols were in accordance with the Animal Ethics Committee of Islamic Azad University.

Extract: Barberry (*Berberis vulgaris*) fruits were obtained from the agriculture garden in Birjand. This is the best barberry native to Iran. These were then washed, chilled to 4°C, and stored. Water extract was prepared from fresh *B. vulgaris* from Iran. Water extract was obtained from distilled water in room temperature. *B. vulgaris* was kept in distilled water (50 g/500 ml) for 72 hours and every 8 hours the preparation was stirred. The final product was administered as a water extract. It should be noted that different concentrations of BV were prepared by diluting the final product with normal saline (0.9 %). The whole juice extracted by the aid of electric juicer and then filtered and stored in clean jars in fridge (0.9% w/v).

Parkinson's disease induction: Parkinson's disease was induced by injection of MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) intraperitoneally (30 mg/kg/b.w./day) for four days [7].

Levodopa-induced dyskinesia (LID): LID was induced by injection of Levodopa (50 mg/kg/day/*i.p.*) for 14 days [8].

Study groups

Animals: In this study 48 male mice were used and randomly divided into six groups (tab. 1).

Abnormal Involuntary Movement Scale (AIMS) test: This preliminary test is used for detection of abnormal movement in mice. Each mouse was observed for 1 minute every 20 minutes. Each mouse was scored from 0 to 4. Zero indicates the absence of symptoms and 4 indicates the most severe form of abnormal

Table 1.

Experiment groups

Study groups
Saline group, (n=8)
MPTP, (n=8)
MPTP+LID,
MPTP+LID+ BV, (5 mg/kg/b.w./*i.p.*), (n=8)
MPTP+LID+ BV, (10 mg/kg/b.w./*i.p.*), (n=8)
MPTP+LID+ BV, (20 mg/kg/b.w./*i.p.*), (n=8)

movement. The rating score was considered as follow: 0: absence of abnormal movement, 1: occasional abnormal movement, 2: frequent abnormal movement, 3: persistent abnormal movement that ceased with sensory stimulation, 4: persistent abnormal movement that was ceased by sensory stimulation [9].

Cylinder test: This test was performed by placing each mouse in a Plexiglas cylinder (20 cm × 30 cm) with the weight-bearing forepaw. Each mouse was observed for 1 minute. Then the number of ipsilateral and contra lateral limb contacts were scored. The final score was calculated with the following formula. Cylinder score: (contra lateral contact – ipsilateral contact)/total contacts × 100 [9].

As the animal moves in an open-top, clear cylinder, its forelimbs activity while rearing against the wall of the cylinder is recorded. Forelimbs use is defined by the placement of the whole forepaws on the wall of the cylinder, which indicates their use for body support. Forelimbs contacts while rearing are scored for each animal in maximum time of one minute. The numbers of impaired and non-impaired forelimbs contacts are calculated as a percentage of total contacts.

Statistics

In this study, an one-way ANOVAs was used to investigate the significant difference of variance among the experimental groups. If the variance showed a significant difference, post hoc Tukey was used to investigate the difference of mean among the groups of study. Data was represented as mean±SEM and $p<0.05$ was considered significant.

RESULTS

Scores of behavioural experiments

Abnormal Involuntary Movement Scale (AIMS) test: Statistical analysis showed that variance has a significant difference among the experimental groups. The AIMS test scores showed that mice in the MPTP + LID group got higher scores than those in the MPTP + LID + BV(10 mg/kg) and MPTP + LID + BV (20mg/kg) - fig.1.

Cylinder Test: Statistical analysis showed that variance has a significant difference among the experimental groups. The cylinder test scores showed that mice in the MPTP + LID group got higher scores than those in the MPTP + LID + BV (10 mg/kg) and MPTP + LID + BV (20 mg/kg) groups - fig. 2.

DISCUSSION

In this study, for the first time the palliative effectiveness of *B. vulgaris* on L-dopa-induced dyskinesia has been shown. This palliative effect was seen at the dosages of 10 and 20 mg/kg.

The *B. vulgaris* (BV) that was used in this study is of a kind that is mainly used as a food supplement [10]. In this sense, using this plant as an adjunct medicine is acceptable and it is free of unwanted side-effects. The usefulness of this plant send back to somewhat 3,000 years ago in the world [11]. In Iran, it dates back 2,500 years. More than 500 genus of BV have been known to have medicinal properties [12]. BV has a wide variety of usages in medicine. It has anti-cancer, anti-inflammatory, antioxidant, antibacterial, analgesic and antinociceptive and hepatic-protective effects [13].

The usefulness of *B. vulgaris* for treating neurodegenerative diseases has been well investigated in previous studies [14]. Jiang *et al.* and Ahmed *et al.* showed that *B. vulgaris* was effective in treating neurodegenerative diseases such as Parkinson's disease, Huntington's and Alzheimer's diseases [14, 15].

The proposed mechanism for this positive effect was also has been investigated. Kim *et al.* showed that the effectiveness of *B. vulgaris* in treating Parkinson's disease is related to the reduction in degeneration of dopaminergic neurons. It also decreases the apoptosis of neurons in the hippocampus [16]. Combination therapy of *B. vulgaris* with captopril also decreased the stiffness of muscles and twitching by improving oxidative stress and acetylcholine esterase activity [17].

In this study, the beneficial effect of *B. vulgaris* on L-dopa-induced dyskinesia was studied. The benefit of *B. vulgaris* on treatment of L-dopa-induced dyskinesia was dose-dependent [2]. At higher dosages, the benefit was more pronounced. Although it might be due to the antioxidant effects, the exact mechanism that underlies this treatment is not clear and needs to be investigated.

L-dopa-induced dyskinesia is long-term side-effect of L-dopa treatment. Many studies have been done to elucidate the molecular mechanism that causes this disability. LID is of particular interest for

many investigations. The proposed mechanism for LID is thought to be related to signal transduction that defect in spiny neurons of striatum [18]. The BV may act through its beneficial effect on the spiny neurons of striatum and related mechanisms.

LID is a vague phenomenon that there is wide range in occurrence of LID in Parkinson's disease patients (9–80%). In animal studies, it has been shown that nigral destruction is necessary for the development of LID [2]. In humans, some factors are necessary for the development of LID. LID will be developed in idiopathic Parkinson's disease, dose, and duration of therapy with Levodopa and possibly some other factors that today is unknown. In this study, to induce LID, MPTP was first administered and then Levodopa treatment was started.

However, LID still remains a problematic issue in clinical practice with many patients appreciating new therapies for this unwanted side-effect. In this study, we proposed BV as a new adjunct therapy for this Levodopa-induced side-effect. This new medication proved itself in AIM Sand cylinder tests where MPTP + Levodopa mice had lower scores when treated with BV compared to the non-treated group.

In this study, we showed that BV exerts its medicinal effect on LID in a dose-dependent manner [19]. The beneficial effect was observed in higher dosages of BV, not at low dosage. The findings confirm that for getting better medication from BV, high dosages of BV should be used in LID. This medicinal effect of BV in a dose-dependent manner might be related to apoptosis [20] and cytotoxicity [21]. However, it is thought that the findings of this study are in accordance with other studies somehow. To answer the question of what is the most effective ingredient of BV, we can say that, based on previous studies, it differs in the various species of BV. Overall, all studies support the importance of isoquinoline alkaloids such as berberine, berbamine, and palmatine [12]. The mediator of the positive effects of BV are related to AMPK and mTOR [22]. However, more studies need to be done to confirm it.

Recent pharmacologic treatment strategies for LID are focused on treatment with monoamine oxidase-B inhibitors, 5-HT agonists and other treatments such as valproate, gabapentin, zonisamide, levetiracetam, topiramate and ACR325 (odopidine).The most useful drugs in this regard are mGluR antagonists and partial DA agonists [23]. However, effective treatment for LID has not yet been confirmed. Like other diseases, it is advisable to use herbal medicine in current practice as an adjunct. LID hampers the quality of life of many Parkinson's patients. Since many patients are looking for an effective treatment for LID, BV will bring successful treatment for such patients,

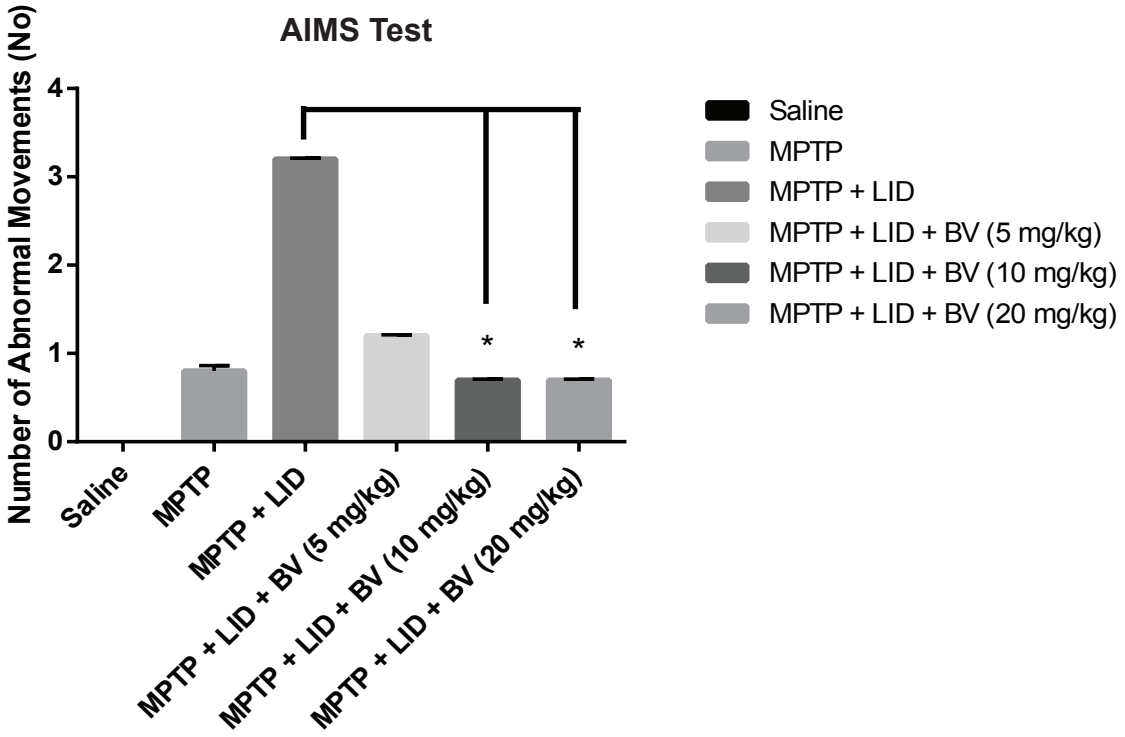

Figure 1.

Scores of different groups of mice fort he AIMS test. Data are represented as mean ±SEM; $p<0.05$ has been considered as significant (n=8)

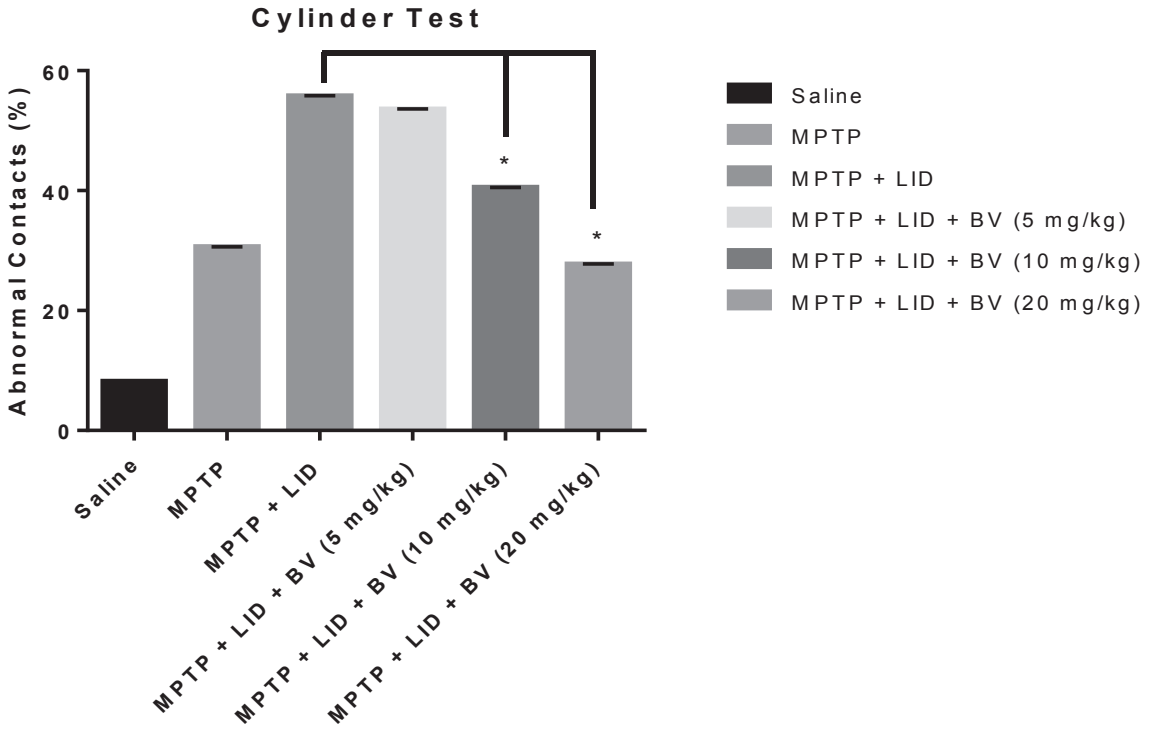

Figure 2.

Scores of different groups of mice for the cylinder test. Data are represented as mean ± SEM; $p<0.05$ has been considered as significant (n=8)

especially, considering that LID will eventually develops in most Parkinsonism patients at later stages of the disease. BV is inexpensive and easy to prepare; in this regard it is also a good candidate and proper alternative for many Parkinson's patients.

CONCLUSION

In this study, we observed that BV had a therapeutic effect for LID in Parkinson's disease. The effective treatment occurred at doses of 10 and 15 mg/kg/b.w. and did not occur at a dose of 5 mg/kg/b.w. So, a higher concentration of the active substance of the extract is recommended for treatment. Overall, this study promises a novel and effective treatment for treating LID in Parkinson's disease patients.

Conflict of interest: Authors declare no conflict of interest.

REFERENCES

1. Gelb DJ, Oliver E, Gilman S. Diagnostic criteria for Parkinson disease. Arch Neurol 1999; 56(1):33-9.

2. Fisone G, Bezard E. Molecular mechanisms of L-DOPA-induced dyskinesia. Int Rev Neurobiol 2011; 98:95-122. doi: http://dx.doi.org/10.1016/B978-0-12-381328-2.00004-3

3. Tomlinson CL, Stowe R, Patel S, Rick C, Gray R, Clarke CE. Systematic review of levodopa dose equivalency reporting in Parkinson's disease. Movement Dis 2010; 25(15):2649-53. doi: http://dx.doi.org/10.1002/mds.23429

4. Luginger E, Wenning G, Bösch S, Poewe W. Beneficial effects of amantadine on L-dopa-induced dyskinesias in Parkinson's disease. Movement Dis 2000; 15(5):873-8.

5. Calixto J. Efficacy, safety, quality control, marketing and regulatory guidelines for herbal medicines (phytotherapeutic agents). Brazilian J Med Biol Res 2000; 33(2):179-89.

6. Rahimi-Madiseh M, Lorigoini Z, Zamani-Gharaghoshi H, Rafieian-Kopaei M. *Berberis vulgaris*: specifications and traditional uses. Iran J Basic Med Sci 2017; 20(5):569-87. doi: http://dx.doi.org/10.22038/IJBMS.2017.8690

7. Schober A. Classic toxin-induced animal models of Parkinson's disease: 6-OHDA and MPTP. Cell

Tiss Res 2004; 318(1):215-24. doi: http://dx.doi.org/10.1007/s00441-004-0938-y

8. Lee CS, Cenci MA, Schulzer M, Björklund A. Embryonic ventral mesencephalic grafts improve levodopa-induced dyskinesia in a rat model of Parkinson's disease. Brain 2000; 123(7):1365-79.

9. Paquette MA, Anderson AM, Lewis JR, Meshul CK, Johnson SW, Berger SP. MK-801 inhibits L-DOPA-induced abnormal involuntary movements only at doses that worsen parkinsonism. Neuropharmacology 2010; 58(7):1002-8. doi: http://dx.doi.org/10.1016/j.neuropharm.2010.01.003

10. Ardestani SB, Sahari MA, Barzegar M, Abbasi S. Some physicochemical properties of Iranian native barberry fruits (abi and poloei): *Berberis integerrima* and *Berberis vulgaris*. J Food Pharm Sci 2013;1(3). doi: http://dx.doi.org/10.15835/nbha46211111

11. Fabricant DS, Farnsworth NR. The value of plants used in traditional medicine for drug discovery. Environ Health Persp 2001; 109(Suppl 1):69.

12. Imanshahidi M, Hosseinzadeh H. Pharmacological and therapeutic effects of *Berberis vulgaris* and its active constituent, berberine. Phytother Res 2008; 22(8):999-1012. doi: http://dx.doi.org/10.1002/ptr.2399

13. El-Wahab AEA, Ghareeb DA, Sarhan EE, Abu-Serie MM, El Demellawy MA. *In vitro* biological assessment of *Berberis vulgaris* and its active constituent, berberine: antioxidants, anti-acetylcholinesterase, anti-diabetic and anticancer effects. BMC Complem Alt Med 2013; 13(1):218. http://dx.doi.org/10.1186/1472-6882-13-218

14. Jiang W, Li S, Li X. Therapeutic potential of berberine against neurodegenerative diseases. Sci China Life Sci 2015; 58(6):564-9. http://dx.doi.org/10.1007/s11427-015-4829-0

15. Ahmed T, Abdollahi M, Daglia M, Nabavi SF, Nabavi SM. Berberine and neurodegeneration: A review of literature. Pharmacol Rep 2015; 67(5):970-9. doi: http://dx.doi.org/10.1016/j.pharep.2015.03.002

16. Kim M, Cho K-H, Shin M-S, Lee J-M, Cho H-S, Kim C-J, et al. Berberine prevents nigrostriatal dopaminergic neuronal loss and suppresses hippocampal apoptosis in mice with Parkinson's disease. Intern J Mol Med 2014; 33(4):870-8. doi: http://dx.doi.org/10.3892/ijmm.2014.1656

17. Salar F, Ziai S, Nasri S, Roghani M, Kamalinejad M. Neuroprotective effect of aqueous extract of *Berberis vulgaris* L. in a model of Parkinson's disease in rat. J Med Plants 2010; 9(36):24-216.

18. Thanvi B, Lo N, Robinson T. Levodopa-induced dyskinesia in Parkinson's disease: clinical features, pathogenesis, prevention and treatment. Postgrad Med J 2007; 83(980):384-8. doi: http://dx.doi.org/10.1136/pgmj.2006.054759

19. Ming M, Sinnett-Smith J, Wang J, Soares HP, Young SH, Eibl G, et al. Dose-dependent AMPK-dependent and independent mechanisms of berberine and metformin inhibition of mTORC1, ERK, DNA synthesis and proliferation in pancreatic cancer cells. PloS ONE 2014; 9(12):e114573. doi: http://dx.doi.org/10.1371/journal.pone.0114573

20. Wang N, Feng Y, Zhu M, Tsang CM, Man K, Tong Y, et al. Berberine induces autophagic cell death and mitochondrial apoptosis in liver cancer cells: the cellular mechanism. J Cell Biochem 2010; 111(6):1426-36. doi: http://dx.doi.org/10.1002/jcb.22869

21. Rad SZK, Rameshrad M, Hosseinzadeh H. Toxicology effects of *Berberis vulgaris* (barberry) and its active constituent, berberine: a review. Iran J Basic Med Sci 2017; 20(5):516. doi: http://dx.doi.org/10.22038/ijbms.2017.8676

22. Liu C-S, Zheng Y-R, Zhang Y-F, Long X-Y. Research progress on berberine with a special focus on its oral bioavailability. Fitoterapia 2016; 109:274-82. doi: http://dx.doi.org/j.fitote.2016.02.001

23. Bargiotas P, Konitsiotis S. Levodopa-induced dyskinesias in Parkinson's disease: emerging treatments. Neuropsych Dis Treat 2013; 9:1605. doi: http://dx.doi.org/10.2147/NDT.S36693

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