Available online at www.scholarsresearchlibrary.com

Scholars Research Library

Der Pharmacia Lettre, 2016, 8 (10):136-139
(http://scholarsresearchlibrary.com/archive.html)

Der Pharmacia Lettre
Scholars Research Library
ISSN 0975-5071
USA CODEN: DPLEB4

Effects of *Pistacia vera* methanolic extract against *Leishmania major* promastigotes

Hossein Mahmoudvand[1] and Farzad Doostishoar[2*]

[1]*Department of Parasitology and Mycology, School of Medicine, Razi Herbal Medicines Research Center, Lorestan University of Medical Sciences, Khorramabad, Iran*
[2]*Kerman Medical Students Research Committee, Kerman University of Medical Sciences, Kerman, Iran*

ABSTRACT

Here, we evaluated the in vitro leishmanicidal effects of Pistacia vera methanolic extract against Leishmania major *promastigotes. The leishmanicidal effects of extract on L.* major *promastigotes were performed using MTT method. The values of IC_{50} were determined using SPSS software. The findings indicated that P. vera methanolic was significantly inhibited promastigote of L. major based on a concentration and time dependent form. The measured IC_{50} values for extract and glucantime were 31.15 µg/mL and 44.6 µg/mL, respectively. The findings indicated leishmanicidal activity of P. vera methanolic on L. major promastigotes. However, further studies will be required to approve these results by checking in the animal models as well as volunteer human.*

Key words: Promastigote, *Leishmania major*; Medicinal plants; Cutaneous leishmaniasis, *P. vera*

INTRODUCTION

Leishmania major which caused zoonotic cutaneous leishmaniasis (ZCL) affects approximatly more than 1 million people annually around the world [1, 2]. Nowadays, chemotherapy with pentavalent antimony components including Glucantime and Pantostame is the preferred treatment for ZCL [3, 4]. However, reviews have reported that these chemical agents because of some problems such as different efficacy, adverse side effects and emergence of resistance [4, 5]. Since ancient times, humans have been applied natural products and plant extracts for treatment of various diseases because of less side effects, low cost and high availability [6]. One of these interesting plants which widely used in Iranian folk medicine is *Pistacia vera* [7]. Based on the previous studies *P. vera* has various pharmacologial properties such as anti-inflammatory, antinociceptive, antiatherogenic, and hypoglycemicones, as well as antibacterial, antiviral, antifungal, and antiparasitic activities against some microbial pathogenic strains [7-9]. Here, we aim to explore the *in vitro* antileishmanial effect of *P. vera* methanolic extract on *L. major* promastigotes.

MATERIALS AND METHODS

Parasite strain

Standard strain of *L. major* (MRHO/IR/75/ER) was kindly obtained from the Laboratory for Leishmaniasis, Tehran University of Medical Sciences, Tehran, Iran, and then cultured in RPMI-1640, supplemented with penicillin (100 IU/mL), streptomycin (100 µg/mL), and 15% heat-inactivated fetal calf serum (FCS) (10, 11).

Plant materials

The *P. vera* leaves were collected from suburbs of Kerman city, Iran, in August 2014. The identity was confirmed by a botanist and voucher sample was recorded at the Herbarium of Department of Pharmacognosy, Kerman University of Medical Science, Kerman, Iran.

Preparing the methanolic extract
The materials were extracted by methanol (80%) for 72 h at 21°C using the percolation method. Then, it was passed via filter paper (Sigma, Germany) to delete plant debris. Finally the isolated extract concentrated in vacuum at 50 °C using a rotary evaporator (Heidolph, Germany) and stored at -20 °C, until use [12-15].

Lesihmanicidal activity
Here, MTT (Sigma-Aldrich, USA) assay was applied to investigate the antileishmanial effect of *P. vera* methanolic extract against *L. major* promastigote form [16, 17]. Breifly, 100 µL of the promastigotes (10^6cells/mL) in logarithmic phase was added to a 96-well plate. Then, 100 µL of various concentrations (0-100 µg/mL) of extract and MA (Aventis, France) was putted to wells and keep at 25°C ± 1°C for 72 hours. Then, 10 µL of MTT solution (5 mg/mL) was putted to wells and keep at 25°C for 4 hours. Finally, absorbance was measured by an ELISA reader (BioTek-ELX800) at 490 nm. 50% inhibitory concentrations (IC_{50} values) were also determined using SPSS software.

Statistical analyses
All the experiments were repeated in triplicate. We used SPSS software, ver. 17, (SPSS Inc., Chicago) for data entry and statistical analysis and differences between the groups were determined using one-way analysis of variance (ANOVA) test. Moreover, to compare IC_{50} values of the groups, *t*-test was performed. *P*-value of less than 0.05 was considered statistically significant [18-20].

RESULTS AND DISCUSSION

Antileishmanial effects
Based on the obtained results, extract of *P. vera* was significantly inhibited promastigote of *L. major* according to a concentration and time dependent form; indicating that with increasing of time and concentration, extract of *P. vera* demonstrated higher leishmanicidal effects compared with control group. The measured IC_{50} values for extract and glucantime were 31.15 µg/mL and 44.6 µg/mL, respectively.

Based on the World Health Organization (WHO) reports, majority of the population around the world used the folk medicine for remedy requirements. Humans from last centuries have been applied plants materials to treat a wide spectrum of diseases such as infectious ones [4, 21]. Reviews have reported different therapeutic effects of *P. vera* in traditional and modern medicines such as antimicrobial, anti-inflammatory, antinociceptive, antioxidant, anti-hepatic ischemia, neuro-protective [7,8]. Our fundings revealed that extract of *P. vera* was significantly inhibited promastigote of *L. major* according to a concentration and time dependent form. The measured IC_{50} values for extract and glucantime were 31.15 µg/mL and 44.6 µg/mL, respectively. Previously we have reported that essential oil of *P. vera* significantly ($P<0.05$) suppressed the growth of *L. tropica* amastigotes based on a dose-dependent response; whereas the IC_{50} value was 23.6 µg/ml against promastigotes [9]. Orhan et al. [21] reported that *P. vera* branch extract at 4.8 µg/mL significantly suppressed (77.3%) the growth of *L. donovani*, whereas the extract revealed potent efficacy on *Plasmodium falciparum*. Orhan et al. [21] also demonstrated that the IC_{50} values of extracts were calculated as 2.3 µg/mL for the amastigotes of *L. donovani* grown in axenic culture and as 3.65 µg/mL for *Plasmodium falciparum.*

Reviews have demonstrated the presence of terpenoid, flavonoids, tannins, and phenols in the phytochemical screenings of the *P. vera* methanolic extract [22, 28]. Previous studies have shown various antimicrobial effects of these constituents particularly terpenoid components such as antibacterial, antifungal, and antiparasitic activities [23-35]. Thus, we can conclude that these components in *P. vera* methanolic extract could be responsible for its antileishmanial activity; while their exact mechanism of action is not clear. However, several studies have shown that [36-47] have reported that some terpenoid can diffuse into pathogens and damage cell memberane structures.
In the case of cytotoxicity effects of *P. vera*, Mahmoudvand et al (2015) exhibited that *P. vera* essential oil had no significant cytotoxicity in J774 cells on *in vitro* [9]. These results showed *P. vera* is safe for mammalian cells.

CONCLUSION

The findings indicated leishmanicidal activity of *P. vera* methanolic on *L. major* promastigotes. However, further studies will be required to approve these results by checking in the animal models as well as volunteer human.

REFERENCES

[1] World Health Organization. Control of the Leishmaniasis. Geneva: WHO (Technical Report Series 949) **2010**; 5–12.

[2] Mahmoudvand, H., Ezzatkhah, F., Sharififar, F., Sharifi, I., & Dezaki, E.S. (**2015a**). *Korean J Parasitol* **2015**, 53, 21–27.
[3] Mahmoudvand H, Ayatollahi Mousavi SA, Sepahvand A, Sharififar F, Ezatpour B, Gorohi F, Saedi Dezaki E, Jahanbakhsh S. *ISRN. Pharmacol* **2014**, 602436.
[4] Desjeux P. *Comp Immunol Microbiol Infect Dis.* **2004**; 27: 305–318.
[5] Croft SL, Sundar S, Fairlamb AH. *Clin Microb Rev*. **2006**; 19(1):11–26.
[6] Rocha LG, Almeida JR, Macedo RO, Barbosa-Filho JM. *Phytomedicine* **2005**; 12: 514-535.
[7] Bozorgi M, Memariani Z, Mobli A, Surmaghi MSH, Shams-Ardekani MR, Rahimi R. *The Scientific World Journal*, vol. **2013**, Article ID 219815, 33 pages, 2013.
[8] Ezatpour B, Saedi Dezaki E, Mahmoudvand H, Azadpour M, Ezzatkhah F. *Evid Based Complement Alternat Med*. **2015,** 149707.
[9] Mahmoudvand, H., Saedi Dezaki, E., Ezatpour, B., Sharifi, I., Kheirandish, F., & Rashidipour, M. *Planta Medica* **2016**, 82(4), 279-284.
[10] Mahmoudvand H, Shakibaie M, Tavakoli R, Jahanbakhsh S, Sharifi, I. *Iran J Parasitol* **2014**, 9(4), 452-460.
[11] Mahmoudvand H, Tavakoli R, Sharififar F, Minaie K, Ezatpour B, Jahanbakhsh S, Sharifi I. *Pharm Biol* **2014**, 4, 1–6.
[12] Rezaeifar R, Ayatollahi Mousavi SA, Mehrabani M, Sepahvand A. *Der Pharm Chem* **2016**; 8(6)l: 140-142.
[13] Rezaeifar M, Behfarnezhad M, Moradi M, Mehrabani M, Mahmoudvand H. *Der Pharmacia Lettre*. **2016**;8(6): 110-112.
[14] Mahmoudvand H, Fasihi Harandi M, Shakibaie M, Aflatoonian MR, Makki MS, Jahanbakhsh S. **2014**. *Int J Surg*. 12, 399–403.
[15] Mahmoudvand H, Asadi A, Harandi MF, Sharififar F, Jahanbakhsh S, Dezaki ES. *Iran J Basic Med Sci*. **2014**, 17(12), 1001-6.
[16] Mahmoudvand H, Sharififar F, Saedi Dezaki E, Ezatpour B, Jahanbakhsh S. Fasihi Harandi M. *Iranian J Parasitol*. **2014**, 9(4), 26-34.
[17] Mahmoudvand H, Sharififar F, Sharifi I, Ezatpour B, Fasihi Harandi M, Makki MS, Jahanbakhsh S. *Iran J Parasitol* **2014**, 9, 28–36.
[18] Tavakoli Kareshk A, Keyhani A, Mahmoudvan d H, Tavakoli Oliaei R, Asadi A, Andishmand M, et al. *Iran J Parasitol*. **2015**, 10 (4), 625-631.
[19] Hosseinzadeh H, Khoshdel M, Ghorbani M. *J Acupunct Meridian Stud* **2011**, 4, 242–247.
[20] Mahmoudvand H, Saedi Dezaki E, Kheirandish F, Ezatpour B, Jahanbakhsh S, Fasihi Harandi M**.** *Korean J Parasitol.* **2014, *52***, *653-659.*
[21] Orhan I, Aslan M, Sener B, Kaiser M, Tasdemir D. *Phytomedicine* **2006**; 13: 735-739.
[22] Mahmoudvand H, Kheirandish F, Ghasemi Kia M, Tavakoli Kareshk A, Yarahmadi M. *Nat Prod Res*. **2015**, 7, 1-4.
[23] Mahmoudvand H, Mirbadie SR, Sadooghian S, Fasihi Harandi M, Jahanbakhsh S, Saedi Dezaki E. *Journal of Essential Oil Research*, **2016**; DOI:10.1080/10412905.2016.1201546
[24] Badparva E, Fallahi Sh, Sepahvand A, Pournia Y, Rashnoo ShM. *Pak J Biol Sci*. **2009**, 12(17), 1212-6.
[25] Mahmoudvand H, Fallahi S, Mahmoudvand H, Shakibaie M, Harandi MF, Dezaki ES. *J Invest Surg*. **2015**, 18, 1-7.
[26] Mahmoudvand H, Ziaali N, Aghaei I, Sheibani V, Shojaee S, Keshavarz H, Shabani M. *Pathogen Glob Health*. **2015**, 109(8), 369-76.
[27] Jahanbakhsh S, Azadpour M, Tavakoli Kareshk A, Keyhani A. Mahmoudvand H. *J Parasit Dis* **2015,** DOI 10.1007/s12639-015-0670-4.
[28] Haidari M, Azargoon A, Mahmoudvand H, Almasi V, Pournia S, Shams Khorramabadi M. *Trauma Mon*. **2014**, 19 (2), e13523.
[29] Rezaeifar M, Mahmoudvand H, Amiria M. *Der Pharma Chemica*, **2016**, 8 (5), 243-249.
[30] Azadpour M, Nowroozi J, Goudarzi GR, Mahmoudvand H. *Trop Biomed*. **2015**; 32(1):109-15.
[31] Vahabi S, Nadri S, Izadi F. *Pak J Pharm Sci* **2014**, 27 (5), 1203-1207.
[32] Mahmoudvand H, Sepahvand A, Jahanbakhsh S, Ezatpour B, Ayatollahi Mousavi SA. *J Mycol Med*. **2014**, 24(4), 155-61.
[33] Teimori H, Sabzi, F, Hassani V, Nadri S, Mahmoudvand H. *Acta Medica Iranica* **2007**, 47(3), 227-232.
[34] Mahmoudvand H, Ziaali N, Aghaei I, Sheibani V, Shojaee S, Keshavarz H, Shabani M. Pathogen Glob Health. 109(8):369-76.
[35] Ahmadinejad M, Aliepour A, Anbari K, Kaviani M, Ganjizadeh H, Nadri S, et al. *Indian J Surg* **2013**, 1-4.
[36] Sikkema J, de Bont JA, Poolman B. *Microbiol Mol Biol Rev* **1995**; 59: 201-222.
[37] Mahmoudvand H, Jahanbakhsh S, Doostishoar F. *Der Pharma Chemica*, **2016**, **8**(8):82-85.
[38] Mahmoudvand H, Ziaali N, Ghazvini H, Shojaee S, Keshavarz H, Esmaeeilpour K, Sheibani V. *Inflamation*. **2016**, 39(1), 405-412.

[39] Mahmoudvand H, Saedi Dezaki E, Soleimani S, Baneshi MR, Kheirandish F, Ezatpour B, Zia-ali N. *Parasite Immunol*. **2015**, 37(7), 362-367.
[40] Saedi Dezaki E, Mahmoudvand H, Sharififar F, Fallahi S, Monzote L, Ezatkhah F. (**2015**). *Pharm Biol*. **2015**, 8, 1-7.
[41] Mahmoudvand H, Sharififar F, Rahmat MS, Tavakoli R, Dezaki ES, Jahanbakhsh S, Sharifi I. *J Vector Borne Dis*. **2014**, 51(4), 294-9.
[42] Kayedi MH, Lines JD, Haghdoost AA. *Acta Tropica* **2009**; **111**:192-196.
[43] Mohammadi M, Shokouh Amiri MR, Sepahvand A, Roodbar Mohammadi SH, Shadzi SH, Mirsafaei H, Noor Shargh R. *J Isfahan Med School* **2009**, 27 (100), 582-588.
[44] Mahmoudvand H, Kheirandish F, Saedi Dezaki E, Shamsaddini S, Fasihi Harandi M. Biomedicine & Pharmacotherapy 82 (**2016**) 393–398
[45] Kayedi MH, Kaur H, Haghdoost AA, Lines JD. *Annals of Tropical Medicine and Parasitology*, **103**: 1-6 (**2009**).
[46] Kheirandish F, Delfan B, Mahmoudvand H , Moradi M , Ezatpoura B, Ebrahimzadehc F, Rashidipour M. *Biomedicine & Pharmacotherapy* **2016**; 82: 208–215.
[47] Mahmoudvand H, Nadri S, Jahanbakhsh S, Rezaeifar M. *Der Pharma Chemica*. **2016**; 8(6): 91-95.