



REVIEW: A Review of the Effects of Herbal Medicines on Leishmaniasis

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ABSTRACT

Leishmaniasis is a tropical mistreated sandfly-borne contagion caused by hemoflagellate protozoa of the *Leishmania* species and it is endemic in many countries such as Iran, Afghanistan, Syria, Saudi Arabia, Brazil, and Peru. After malaria, leishmania spp. causes the highest rates of mortality and morbidity. Several major risk factors are involved in the spread of leishmaniasis such as drug resistance, environmental changes, HIV epidemic, inadequate vector control and migration of non-immune individuals to endemic areas. Leishmaniasis is a disease with diverse clinical manifestations that depends on both infecting species of *Leishmania* and the immune response of the host. Different forms of the disease include cutaneous, mucocutaneous and visceral forms. The most dangerous form is Visceral Leishmaniasis (VL) which can be fatal among untreated patients. The availability of the inadequate number of antileishmanial chemotherapeutic compounds, high-cost treatment, rising drug resistance as well as severe toxicities of the drugs obscures the treatment of VL. Many investigations showed antileishmanial activity of herbal extracts or chemical derivatives from natural sources *in vitro* against promastigote and amastigote forms or *in vivo* against *Leishmania*-infected animals. A review of related studies revealed that herbal extracts demonstrating antileishmanial activities *in vivo* or *in vitro* alone or combined with suggested drugs seem to confirm their use in folk medicine. Additionally, the antileishmanial activity of more than a hundred plants have been recognized in this regard. It is worth noting that plants are notable sources of medicine production, because of their long association with parasites.

Introduction

L Leishmaniasis is a common disease among humans and animals and it is caused by a protozoan parasite of the genus *Leishmania* (1). *Leishmania* is a protozoan of the trypanosomatid family of the genus *Sarcomastigophora* which has two distinct stages in its life cycle. The promastigote form is flagellated and motile and reproduces in the intestine of the sandfly. The amastigote form is immobile and settles inside the mammalian host's macrophages

(2). Leishmaniasis is endemic in many tropical countries and in Iran it is known as Oriental ulcer, too (1).

Leishmaniasis is transmitted through infected female sandflies including *Phlebotomus* and *Lutzomyia*. The disease is endemic in more than 98 countries and an estimated 350 million people are at risk of infection. The overall prevalence is 12 million and the annual incidence is nearly 2 - 2.5 million. In most countries, the number of cases is likely

to be underestimated because cases are not detected and reporting is not obligatory. Depending on the infected species, infection with *Leishmania* parasites can lead to three main clinical manifestations. The first form is named localized Cutaneous Leishmaniasis (3) with single or multiple skin lesions, satellite lesions, or nodular lymphangitis (4). It is the most common form and causes skin wounds, lasting lesions with a serious disability. The second form is Mucocutaneous Leishmaniasis (MCL), which causes total or partial damage of nose, mouth and throat mucous. The third one is systemic Visceral Leishmaniasis (VL) which involves internal organs such as liver, spleen and bone marrow. It can be fatal among untreated individuals (4, 5).

Different classifications are considered for leishmaniasis. According to geographical classification, it is divided into New World and Old World disease. Old World forms of leishmaniasis are endemic in Africa, Asia, the Middle East, and the Mediterranean. These are transmitted through the blood meal of *Phlebotomus* sandflies. While the New World form is transmitted mainly by *Lutzomyia* flies. In this regard, Texas through South America is a leishmaniasis-endemic region (6).

In the New World, CL is caused by many species of the subgenera *Leishmania* and *Viannia* parasites, primarily *L. amazonensis*, *L. braziliensis*, *L. guyanensis*, *L. mexicana* and *L. panamensis*, whereas in the Old World, CL is just caused by five species of *Leishmania* including *L. aethiopica*, *L. donovani*, *L. infantum*, *L. major* and *L. tropica* (7). CL has been clinically observed in Iran in two forms: rural (wet wound) and urban (dry wound). Rural CL is a common disease of humans and animals and is called Zoonotic Cutaneous Leishmaniasis (ZCL). Urban CL is known as Anthroponotic Cutaneous Leishmaniasis (ACL). The causative agent of urban CL is *L. tropica* (8).

Based on the World Health Organization (WHO) report, in 2020 over 85% of new CL cases were observed in 10 countries: Afghanistan, Algeria, Brazil, Colombia, Iraq,

Libya, Pakistan, Peru, the Syrian Arab Republic and Tunisia. While, over 90% of MCL cases were reported in Bolivia, Brazil, Ethiopia and Peru. Concerning VL, more than 90% of new cases were detected in 10 countries: Brazil, China, Ethiopia, Eritrea, India, Kenya, Somalia, South Sudan, Sudan and Yemen (5).

Currently, the most important drug used to treat various clinical forms of leishmaniasis is the 5-valent antimony compounds, which include glucantime and pentostam. Following the administration of these drugs, there have been reports of significant side effects. It is pointing that topical treatments, heat therapy and cold therapy are used to treat CL. Today, due to the resistance of genus *Leishmania* to common antibiotics, replacing them with newer antibiotics is desirable. Likewise, the tendency to use herbal medicinal compounds is so widespread these days. In this regard, 25% of the currently used drugs are derived from plant sources (9).

The point is that effective treatment of CL with glucantime usually requires injection at the site of the lesion, and since the lesions are mainly seen on hands or face, the injection of the drug is painful. Concerning its effect on the parasite, glucantime can destroy the person's normal cells and it is one of the most important side effects of glucantime treatment. In most cases wounds do not cause a serious problem for the patient and recover spontaneously, but for a variety of reasons such as the length of the wound recovery, the deformity of the scar, and the possibility of secondary infections at the site of the lesion, providing tolerable treatment without side effect seems reasonable. Thus, the use of herbal products and native plants of endemic areas that provide a rich source of anti-leishmaniasis compounds is one of the important goals of the WHO and other health institutions in the world and Iran. In fact, medicinal plants have fewer side effects in comparison with chemical drugs (10).

Methods

In this non-systematic review, the data were

gathered via searching keywords and phrases such as herbal medicines, therapeutic plants, Leishmania protozoa and Leishmaniasis and a combination of those in PubMed, Science Direct, ELSEVIER, Google Scholar, Embase and SID (Scientific Information Database) from 2000 to 2020. The results of the related studies were used in this investigation.

Literature review

Review of related studies revealed that nutritional value and medicinal plants have been considered for the control and treatment of diseases due to their various compounds, including antioxidants. The most important benefits of herbal medicines are their low cost, low incidences of serious side effects and their respectable effectiveness. Unfortunately, the effectiveness of the herbal compounds has only been shown in laboratory studies and they are still in the early stages of clinical trials and studying their results in clinical practice is a neglected issue. It is estimated that plant products are directly or indirectly involved in the production of about 25% of drugs (11).

Several studies have shown that different plant species have inhibitory activity against certain types of parasites such as *L. major*. Some plants' oil has regulatory effects on the immune system, which makes them useful in the treatment of leishmaniasis. In this regard, several novel compounds have proved leishmanicidal activity. Besides, a vast number of herbal extracts, such as alkaloids, chalcones, phenolics and terpenes are presented for treatment (12). The antileishmanial activity of some herbal extracts has been attributed to flavonoids. Flavonoids are a group of polyphenolic compounds found naturally in fruits and vegetables and are known as antioxidants and anti-cancer drugs with significant protective effects against membrane damage. Flavonoids are able to complex with the parasite's cell wall to affect the processes of cell attachment and inhibit parasitic growth (13).

Even though the use of therapeutic plants in leishmaniasis treatment has been interrupted

by unknown mechanisms, researchers are looking for isolating pure compounds from herbal for immunomodulatory properties, antileishmanial activity or to decrease drug toxicity (14-16). Thus, the current pattern of drug discovery has failed to meet the treatment needs for high-priority diseases in developing countries. In fact, leishmaniasis is a good example that almost no new effective drugs have been produced in the last 70 years (17). Concerning health issues, several studies have investigated the effect of some herbal extracts on leishmaniasis (18-21).

In this realm, García et al, investigated the antileishmanial action of 21 species of plants. They collected plants and screened their hydroalcoholic extracts against promastigotes and amastigotes of *L. amazonensis*. Their toxicity was also assayed against peritoneal macrophages from BALB/c mice. Five extracts showed significant growth inhibitory activity against promastigote form. Only the extracts from *Bidens pilosa* L. (Asteraceae) and *Punica granatum* L. (Punicaceae) inhibited the growth of intracellular amastigotes, with IC₅₀ values of 42.6 and 69.6 µg/mL, respectively. In addition, low toxicity on macrophages from BALB/c mice was observed too (22). In the same vein Zahir et al, evaluated the antileishmanial activity of acetone and methanol leaf extracts of *Anisomeles malabarica*, *Ocimum basilicum*, the flower of *Gloriosa superba*, leaf and seed of *Ricinus communis* against *L. donovani*. Results demonstrated that leaf methanol extracts of *A. malabarica*, and *R. communis* showed good antileishmanial activity and could play an important role in herbal formulations for the treatment of VL. In **Table 1** and **Table 2**, main outcomes of some related studies have been highlighted (23).

Conclusion

Leishmaniasis includes a group of diseases caused by numerous species of *Leishmania* and shows a variety of clinical symptoms. Besides, contagions due to protozoa of the genus *Leishmania* are a main worldwide health problem, with high endemicity in dev-

Table 1. Related studies in world

Authors	Year	Medicine source	Main Findings
Le Pape, et.al (24)	2000	<i>Pachymatisma johnstonii</i> ,	High activity of isolated sponge glycoprotein against <i>L. donovani</i> , <i>L. braziliensis</i> and <i>L. mexicana</i> and aphidicolin <i>in vitro</i> was reported.
		<i>nigrospora sphaerica</i>	The growth of promastigotes and amastigotes of <i>L. donovani</i> was inhibited by a fungal metabolite.
Plock, et.al (25)	2001	<i>Yucca filamentosa</i>	Ethanol extract of <i>Yucca filamentosa</i> had potent activity against <i>L. amazonensis</i> at a concentration of 5 mg/mL.
Lamidi, et al. (26)	2005	<i>Polyalthia suaveolens</i>	<i>L. infantum</i> showed the highest effect to methanolic extracts from the leaves of <i>Polyalthia suaveolens</i> , <i>Dioscorea preussii</i> , <i>Augouardia letestui</i> and stem bark of <i>Cola lizae</i> plants (IC50 < 5 µg/mL).
		<i>Dioscorea preussii</i>	
		<i>Augouardia letestui</i>	
Braga, et al. (27)	2007	<i>Vernonia polyanthes</i>	Ethanol extracts of medicinal plants <i>Vernonia polyanthes</i> and <i>Ocimum gratissimum</i> were most active against <i>L. amazonensis</i> at IC50 (4 µg/mL) and <i>L. chagasi</i> at IC50 (71 µg/mL).
		<i>Ocimum gratissimum</i>	
Dutta, A. et al. (28)	2007	<i>Asparagus racemosus</i>	A water-soluble compound, obtained from the fruits of <i>Asparagus racemosus</i> was effective against antimonial-sensitive as well as antimonial-resistant <i>L. donovani</i> promastigotes by inducing apoptosis.
Fokialakis, et al. (29)	2007	<i>Eryngium amorginum</i>	IC50 < 10 µg/mL with no cytotoxicity was observed by treating with dichloromethane extracts of <i>Eryngium ternatum</i> , <i>Origanum Dictamnus</i> , <i>Origanum microphyllum</i> and the methanolic extracts of <i>Eryngium amorginum</i> .
		<i>Eryngium ternatum</i>	
		<i>Origanum Dictamnus</i>	
		<i>Origanum microphyllum</i>	
		<i>Origanum microphyllum</i>	
Mori, et al. (30)	2008	<i>Cordia fragrantissima</i>	The crude extract isolated from <i>Cordia fragrantissima</i> wood had minimal inhibitory concentration of 12.5 µg/mL against <i>L. major</i> promastigotes.
Misra, et al. (31)	2009	<i>Piper betle</i>	Methanolic extract of <i>Piper betle</i> landrace <i>Bangla Mahoba</i> had selective inhibitory effect on promastigote and amastigote forms of Leishmania parasite by persuading apoptosis without toxicity effect on uninfected macrophages.
Mohammad BA. (32)	2011	<i>Aloe vera</i>	The combination of traditional drugs <i>Aloe vera</i> , <i>Euphorbia milli</i> with turmeric and animal fat showed noticeably good antileishmanial activity. It has represented more efficient remedy resulted in wound healing and tissue softening in comparison with glucantime treatment in mice.
		<i>Euphorbia milli</i>	
		animal fat	
Rodrigues, et. al (33)	2011	<i>Syagrus coronata</i>	Outcomes revealed <i>in vitro</i> leishmanicidal activity of aqueous extract of <i>Syagrus coronata</i> on <i>L. amazonensis</i> with minimal inhibitory concentration of 8.3 µg/mL had no cytotoxic effects on mammalian cells.
Rondon, et al. (3)	2011	<i>Aloe vera</i> <i>Coriandrum sativum</i> <i>Ricinus communis</i>	The extract of these plants presented potent activity against promastigote and amastigote forms of Leishmania parasite.
Iqbal, et. al. (34)	2012	<i>Aloe vera leaf</i> <i>Tamarix aphylla bark</i>	<i>Tamarix aphylla</i> bark and <i>Aloe vera</i> leaf had considerable effect on motility rate of <i>L. tropica</i> .
Jain, et al. (35)	2013	<i>Agave americana</i>	<i>Agave americana</i> and <i>Azadirachta indica</i> showed the significant toxicity while <i>Eclipta alba</i> and <i>Piper longum</i> showed the least or negligible toxicity.
		<i>Azadirachta indica</i>	
		<i>Eclipta alba</i>	
		<i>Piper longum</i>	

Kyriazis, et al. (16)	2013	<i>Olea europaea var koroneiki</i>	The oleuropein compound extracted from olive tree decreased the load of parasite in <i>L. donovani</i> infected mice.
Mishra, et al. (36)	2013	<i>Plumbago zeylanica</i>	A naphthoquinone compound, prenyloxy-naphthoquinone extracted from roots of <i>Plumbago zeylanica</i> presented considerable activity against promastigote and amastigote forms of <i>L. donovani</i> .
Lezama-Dávila, et al. (37)	2014	<i>Pentalinon andrieuxii</i>	This investigation represented both immunomodulatory and antileishmanial in vitro activities of hexane extract of <i>Pentalinon andrieuxii</i> (PARE) roots.

Table 2. Related studies in Iran

Authors	Year	Medicine source	Main Findings
Nilforoushzadeh, et.al (38)	2008	<i>Achillea millefolium</i> (yarrow)	The highest efficacy of herbal extracts in reduction of ulcer size was reported for propolis, followed by <i>Achillea millefolium</i> and then <i>Thymus vulgaris</i> .
		<i>Thymus vulgaris</i> (thyme)	
Bonyadian, et al. (39)	2015	<i>Lavender</i>	Findings revealed that the proliferation rate of promastigotes reduced meaningfully after adding plant essential oil. Besides, it indicated the inhibitory effect of growth and lethality of lavender essential oil in vitro on the shape of <i>Leishmania major</i> promastigote.
Eskandari et al. (8)	2016	<i>Medicago lupulina</i>	Alcoholic extract and essential oil of the tested plant had significant anti-leishmaniasis effects in vitro and they can be considered as anti-leishmaniasis medicinal plants.
Nasiri et al. (40)	2016	<i>Black tea</i>	The outcome showed that black tea decoction has a beneficial effect on the elimination of <i>L. major</i> promastigotes in extrinsic conditions.
Bagherian et al. (41)	2017	<i>Thyme</i>	The results of light absorption and IC 50 showed that thyme is effective for the treatment of CL.
Ghaderi et al. (10)	2018	<i>Alpha-pinene compound</i>	Findings revealed the antileishmanial effect of alpha-pinene on <i>L. major</i> promastigotes, in vitro. Moreover, topical ointment of the extract could decrease size of the wounds caused by the parasite, in vivo.
Poursafavi et al. (42)	2018	<i>Olive</i>	The results showed that the aqueous extract at a concentration of 2.5 mg / ml and the hydroalcoholic extract of olive leaf at a concentration of 25 µg / ml on the third day killed all glucantime-sensitive <i>Leishmania tropica</i> amastigotes within macrophages.
Mardani et al. (43)	2020	<i>Cornus mas</i>	The results showed the effect of <i>Cornus mas</i> extract on inhibiting parasite growth influenced by the dose and time of treatment. Besides, all concentrations of the extract were able to reduce wound diameter and parasitic load.

eloping countries. In the present investigation, useful and identified natural products for three forms of leishmaniasis treatment were reviewed. It seems crucial that the antileishmanial activity of herbal extracts to be investigated in all phases of leishmania parasite regardless of the species. The dominant issue is the identification of potent chemical leishmanicidal compounds isolated from natural sources as a step forward in the exploration of antileishmanial drugs. Future perspective in the treatment of leishmaniasis

depends on vaccine development, vector control, screening the effectiveness of treatment and high diagnosis sensitivity. The severity and diversity of leishmaniasis, the inadequacy of antileishmanial medicines and variable reactions in different locations made researchers discover novel antileishmanial compounds including arylimidamide, buparvaquone and nitroquinolines. This can lead to the design of many platforms for the upcoming products of second-generation compounds for the leishmaniasis treatment.

Supplementary improvement in disease control can be achieved by genomic identification of parasites. In addition, information on medicines used for other contagions and new natural extracts might be useful in finding novel advantageous strategies to alleviate this infection.

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Conflicts of Interest

None has been announced.

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References

1. Pirali Kheirabadi K, Hejazi H, Layeghi Ghale Soukhteh S. Cytotoxic effect of essential oil of *Artemisia Siberi* and *Pelargonium roseum* on *Leishmania major* Promastigotes. *Journal of Shahrekord Ununiversity of Medical Sciences*. 2015;17(5): 13-22.
2. Soleimanifard S, Arjmand R, Hejazi SH. Investigation and Comparison of *Leishmania major* Promastigote and Amastigote Protein Content by Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis. *Avicenna Journal of Clinical Medicine*. 2013;20(1):1-8.
3. Rondon FC, Bevilacqua CM, Accioly MP, Morais SM, Andrade-Junior HF, Machado LK, et al. In vitro effect of *Aloe vera*, *Coriandrum sativum* and *Ricinus communis* fractions on *Leishmania infantum* and on murine monocytic cells. *Veterinary parasitology*. 2011;178(3-4):235-40.

4. de Vries HJ, Reedijk SH, Schallig HD. Cutaneous leishmaniasis: recent developments in diagnosis and management. *American journal of clinical dermatology*. 2015;16(2):99-109.
5. WHO. Leishmaniasis: World Health Organization; 2022 [Available from: <https://www.who.int/en/news-room/fact-sheets/detail/leishmaniasis>].
6. Craig G Stark M, FACP, FFTM, RCPS(Glasg), FISTM Telemedicine and Digital Health, CedarBridge Capital Partners. *Leishmaniasis 2020* [Available from: <https://emedicine.medscape.com/article/220298-overview>].
7. WHO. Control of the leishmaniasis. 2010.
8. Gharirvand Eskandari E, Doudi M. Investigation of Antileishmanial Effect of Alcoholic Extract and Essential Oil of Medicinal Plant Leaf Black Alfalfa (*Medicago Lupulina*), on The Number of Clinical Isolates of *Leishmania Major* Promastigotes in Vitro. *The Journal of Shahid Sadoughi University of Medical Sciences*. 2016;24(2):174-84.
9. Fouladvand M, Khorami S, Naeimi B, Fotouhi S, Mohammadi K. Evaluation of Lethal Effect of Curcumin and its Derivatives Against *Leishmania Major* In Vitro. *Iranian South Medical Journal*. 2020;23(2):153-64.
10. Ghaderi A, Kkhadem Eerfan MB, Barati M, Ghaderi S. Evaluation of antileishmanial effect of the plant extract of alpha-pinene(*Pistacia atlantica*) in vitro and in vivo. *Scientific Journal of Kurdistan University of Medical Sciences*. 2018;23(5): 32-44.
11. Moosavi T, Zakavi A, Hosseinivaliki F, Yousef pour M, Fakhar M, Rafiei A, et al. Nutritional Properties of Garlic According to Traditional and Modern Medicine: A Review Study. *Journal of Mazandaran University of Medical Sciences*. 2016;26(139):227-45.
12. Davis AJ, Kedzierski L. Recent advances in antileishmanial drug development. *Current opinion in investigational drugs* (London, England: 2000). 2005;6(2): 163-9.
13. Oryan A. Plant-derived compounds in

treatment of leishmaniasis. Iranian journal of veterinary research. 2015;16(1):1.

14. Das S, Roy P, Mondal S, Bera T, Mukherjee A. One pot synthesis of gold nanoparticles and application in chemotherapy of wild and resistant type visceral leishmaniasis. Colloids and Surfaces B: Biointerfaces. 2013;107:27-34.

15. Sachdeva H, Sehgal R, Kaur S. Studies on the protective and immunomodulatory efficacy of *Withania somnifera* along with cisplatin against experimental visceral leishmaniasis. Parasitology research. 2013;112(6):2269-80.

16. Kyriazis JD, Aligiannis N, Polychronopoulos P, Skaltsounis A-L, Dotsika E. Leishmanicidal activity assessment of olive tree extracts. Phytomedicine. 2013;20(3-4):275-81.

17. Surur AS, Fekadu A, Makonnen E, Hailu A. Challenges and opportunities for drug discovery in developing countries: the example of cutaneous leishmaniasis. ACS Medicinal Chemistry Letters. 2020;11(11):2058-62.

18. Fournet A, Muñoz V, Roblot F, Hocquemiller R, Cavé A, Gantier JC. Antiprotozoal activity of dehydrozaluzanin C, a sesquiterpene lactone isolated from *Munnozia maronii* (Asteraceae). Phytotherapy Research. 1993;7(2):111-5.

19. Mohebbali M, Chenari A, Nazari M. The effect of cassia fistula extracts on leishmania major ulcers in laboratory mice. Pajoohandeh J. 1999;4(1):9-14.

20. Tadesse A, Gebre-Hiwot A, Asres K, Djote M, Frommel D. The in vitro activity of *Vernonia amygdalina* on *Leishmania aethiopica*. Ethiopian Medical Journal. 1993;31(3):183-9.

21. Tahir AE, Ibrahim AM, Satti GM, Theander TG, Kharazmi A, Khalid SA. The potential antileishmanial activity of some Sudanese medicinal plants. Phytotherapy Research: An International Journal Devoted to Pharmacological and Toxicological Evaluation of Natural Product Derivatives. 1998;12(8):576-9.

22. García M, Monzote L, Montalvo AM, Scull R. Screening of medicinal plants against

Leishmania amazonensis. Pharmaceutical biology. 2010;48(9):1053-8.

23. Zahir AA, Rahuman AA, Pakrashi S, Ghosh D, Bagavan A, Kamaraj C, et al. Evaluation of antileishmanial activity of South Indian medicinal plants against *Leishmania donovani*. Experimental parasitology. 2012;132(2):180-4.

24. Le Pape P, Zidane M, Abdala H, Moré M-T. A glycoprotein isolated from the sponge, *Pachymatisma johnstonii*, has antileishmanial activity. Cell Biology International. 2000;24(1):51-6.

25. Plock A, Sokolowska-Köhler W, Presber W. Application of flow cytometry and microscopical methods to characterize the effect of herbal drugs on *Leishmania* Spp. Experimental parasitology. 2001;97(3):141-53.

26. Lamidi M, DiGiorgio C, Delmas F, Favel A, Mve-Mba CE, Rondi M, et al. In vitro cytotoxic, antileishmanial and antifungal activities of ethnopharmacologically selected Gabonese plants. Journal of ethnopharmacology. 2005;102(2):185-90.

27. Braga FG, Bouzada MLM, Fabri RL, Matos MdO, Moreira FO, Scio E, et al. Antileishmanial and antifungal activity of plants used in traditional medicine in Brazil. Journal of ethnopharmacology. 2007;111(2):396-402.

28. Dutta A, Ghoshal A, Mandal D, Mondal NB, Banerjee S, Sahu NP, et al. Racemoside A, an anti-leishmanial, water-soluble, natural steroidal saponin, induces programmed cell death in *Leishmania donovani*. Journal of medical microbiology. 2007;56(9):1196-204.

29. Fokialakis N, Kalpoutzakis E, Tekwani B, Khan S, Kobaisy M, Skaltsounis A, et al. Evaluation of the antimalarial and antileishmanial activity of plants from the Greek island of Crete. Journal of natural medicines. 2007;61(1):38-45.

30. Mori K, Kawano M, Fuchino H, Ooi T, Satake M, Agatsuma Y, et al. Antileishmanial compounds from *Cordia fragrantissima* collected in Burma (Myanmar). Journal of natural products. 2008;71(1):18-21.

31. Misra P, Kumar A, Khare P, Gupta S, Kumar N, Dube A. Pro-apoptotic effect of the landrace Bangla Mahoba of Piper betle on *Leishmania donovani* may be due to the high content of eugenol. *Journal of medical microbiology*. 2009;58(8):1058-66.
32. Mohammad BA. Antileishmanial effects of traditional herbal extracts against cutaneous leishmaniosis in vivo. *Advances in Environmental Biology*. 2011:3188-96.
33. Rodrigues IA, Alviano DS, Gomes MT, Silva DO, Antoniassi R, Silva AJR, et al. In vitro anti-*Leishmania amazonensis* activity of the polymeric procyanidin-rich aqueous extract from *Syagrus coronata*. *Journal of Medicinal Plants Research*. 2011;5(16):3781-90.
34. Iqbal H. Comparative efficacy of Aloe vera and *Tamarix aphylla* against cutaneous leishmaniasis. *International Journal of Basic Medical Sciences and Pharmacy (IJBMS)*. 2012;2(2).
35. Jain K, Jain NK. Novel therapeutic strategies for treatment of visceral leishmaniasis. *Drug discovery today*. 2013;18(23-24):1272-81.
36. Mishra BB, Gour JK, Kishore N, Singh RK, Tripathi V, Tiwari VK. An antileishmanial prenyloxy-naphthoquinone from roots of *Plumbago zeylanica*. *Natural product research*. 2013;27(4-5):480-5.
37. Lezama-Dávila CM, Pan L, Isaac-Márquez AP, Terrazas C, Oghumu S, Isaac-Márquez R, et al. Pentalinon andriouxii root extract is effective in the topical treatment of cutaneous leishmaniasis caused by *Leishmania mexicana*. *Phytotherapy Research*. 2014;28(6):909-16.
38. Nilforoushzadeh M, Shirani-Bidabadi L, Zolfaghari-Baghbaderani A, Saberi S, Siadat A, Mahmoudi M. Comparison of *Thymus vulgaris* (Thyme), *Achillea millefolium* (Yarrow) and propolis hydroalcoholic extracts versus systemic glucantime in the treatment of cutaneous leishmaniasis in balb/c mice. *J Vector Borne Dis*. 2008;45(4):301-6.
39. Bonyadian M, Hejazi H, Azizi H, Habibian S, Sayahi E. Antileishmania activity of *Levandula officinalis* essence against *Leishmania major* in in vitro media. *Journal of Shahrekord Uuniversity of Medical Sciences*. 2015;17(3):93-101.
40. nasiri v, karimi g, payekari h, motamedi g, rivaz s, norouzi e. In vitro evaluation of the effect of black tea decoction extract on *Leishmania major* promastigotes. *Veterinary Clinical Pathology The Quarterly Scientific Journal*. 2015;8(4 (32) Winter): 621-9.
41. Bagherain A, Hejazi SH, Mirzaei M, Mirzaei H, Mirzaei HR, Masoud Khoy MJ. Effect of thyme plants on *Leishmania amastigotes* in invitro: compared with Amphotericin B. *Med J Tabriz Uni Med Sciences Health Services*. 2017;39(2):6-13.
42. Poursafavi Z, Seyyed Tabaei SJ, Kheirandish F, Mohebbali M, Kamalinezhad M, Vajdian R. Effect of olive leaf aqueous and hydroalcoholic extract on *Leishmania tropica* Glucantime resistance and sensitivity in vitro. *Pejoughesh dar Pezeshki (Research in Medicine)*. 2018;42(1):46-52.
43. Mardani HR, Abdizadeh R, Lori Gooini Z, Khalili B. A study on the effect of hydroalcoholic extracts of *Cornus mas* on *Leishmania major* in vitro condition and wounds in Balb/C mice. *Journal of Medicinal Plants*. 2020;19(74):239-54.