



Revista Brasileira de Farmacognosia

BRAZILIAN JOURNAL OF PHARMACOGNOSY

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Original article

Plant species used in giardiasis treatment: ethnopharmacology and *in vitro* evaluation of anti-*Giardia* activity



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ARTICLE INFO

Article history:

Received 15 December 2013

Accepted 13 April 2014

Keywords:

Ethnopharmacology

Giardia lamblia

Giardicidal activity

In vitro

Medicinal plants

ABSTRACT

The aim of this study was to compile the traditional knowledge about plants used for the treatment of giardiasis, and also to carry out experimental research to evaluate the anti-*Giardia* activity of five species. To reach this objective, 398 interviews were performed using a previously prepared questionnaire, followed by an *in vitro* evaluation of giardicidal potential of hydroalcoholic leaf extracts of *Anacardium occidentale* L., *Chenopodium ambrosioides* L., *Passiflora edulis* Sims, *Psidium guajava* L., and *Stachytarpheta cayennensis* (Rich.) Vahl. Among the interviewed people, 55.53% reported the use of plants to treat diarrhea, the most severe symptom of giardiasis. The results indicated 36 species used by this population for these problems. The use of leaves (72.50%) of a single plant (64.25%) collected from backyards and gardens (44.34%) and prepared by decoction were predominant. The majority of the interviewees (85.52%) attributed their cure to the use of plants. In the experimental tests, all extracts inhibited the growth of *Giardia lamblia* trophozoites in different intensities: *A. occidentale* and *P. guajava* extracts elicited a moderate activity ($250 \leq IC_{50} \leq 500 \mu\text{g/ml}$), *C. ambrosioides* and *S. cayennensis* extracts evoked a high activity ($100 \leq IC_{50} \leq 250 \mu\text{g/ml}$), and *P. edulis* extract showed very high activity ($IC_{50} \leq 100 \mu\text{g/ml}$). This study shows that an ethnopharmacological approach is useful in the selection of plant materials with potential giardicidal activity.

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Introduction

Giardiasis is an intestinal infection caused by the flagellate protozoan *Giardia lamblia* (synonyms: *Giardia intestinalis* and *Giardia duodenalis*), with worldwide distribution, high

prevalence, and significant morbidity (Rocha, 2003; Arani et al., 2008). Data from the World Health Organization report about 400 million new cases of *G. lamblia* infection per year (WHO, 2009). Since giardiasis is not a notifiable disease in Brazil, few prevalence records are available; however, several

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<http://dx.doi.org/10.1016/j.bjp.2014.04.004>

epidemiological studies document infection rates of up to 70.5%, indicating that it is a serious public health problem (Borges et al., 2011).

Clinical manifestations of giardiasis vary, however it's most predominant symptom, diarrhea, occurs in 90% of symptomatic patients. Diarrhea can be acute and self-limiting, or chronic and debilitating, associated with abdominal pain, flatulence, dyspepsia, epigastric pain, nausea, vomiting, steatorrhea, low-lipid stools and fat-soluble vitamin absorption, and weight loss. The poor absorption of fats, carbohydrates, iron, and vitamins (A and B12) can delay physical and mental development, especially in younger individuals (Heresi et al., 2000; Lebwohl et al., 2003; Al-Mekhlafi et al., 2005).

The typical treatment for giardiasis is chemotherapy using one or more drugs, predominantly 5-nitroimidazole derivatives such as metronidazole, a first-line drug. Other nitroimidazoles (secnidazole, tinidazole, and ornidazole), benzimidazoles (albendazole, mebendazole), furazolidone, quinacrine, and benzimidazole derivatives have been used for giardiasis treatment. However, chemotherapeutic agents cause adverse reactions such as gastrointestinal disturbances, nausea, headache, leukopenia and parageusia; and may trigger neurotoxic effects as ataxia, seizures, and vertigo, leading to discontinuation of treatment. Moreover, mutagenic and carcinogenic effects have been observed in laboratory animals (Harris et al., 2001; Campanati and Monteiro-Leal, 2002; Petri-Jr, 2003; Andrade et al., 2010). Because of the side effects of conventional drugs and an increased parasite resistance to treatment, it is necessary to identify new effective and safe agents for the treatment of this infection (Upcroft and Upcroft, 2001; Sangster et al., 2002; Hernández and Hernández, 2009).

Natural resources, especially of plant origin, are important sources of new bioactive products, considering the wide variety and complexity of metabolites with potential therapeutic value (Pinto et al., 2002; Anthony et al., 2005; Gurib-Fakim, 2006; Oliveira et al., 2011).

The assessment of therapeutic potential aimed at developing herbal or phytochemical products requires validation of the plant species through ethnobotanical, ethnopharmacological, chemical, biological, pharmacological, and toxicological studies (Gilani and Rahman, 2005; Macêdo and Oliveira, 2006; Klein et al., 2009). Ethnobotanical and ethnopharmacological studies have been shown to provide important findings in the search for new active products of plant origin, effectively contributing to defining the inclusion and exclusion criteria for the selection of plant species and their subsequent validation (Gilani and Rahman, 2005; Patwardhan, 2005; Albuquerque and Hanazaki, 2006).

Considering the high global prevalence of giardiasis and the need for new therapeutic options, taking into account the ethnopharmacological approach in the research and development of drugs of plant origin, an ethnopharmacological survey was conducted to identify the plant species traditionally used to treat giardiasis in the municipality of São Luís, Maranhão State, Brazil. Afterwards, validation studies for anti-Giardia activities of the plant species were conducted.

Materials and methods

The study was approved by the Ethics Committee of the Federal University of Maranhão under protocol number 23115-012975/2008-43. Prior to data collection, the participants were asked to sign an informed consent form authorizing participation and dissemination of the data collected.

Ethnopharmacological study

Type of study

The first stage of the research was a descriptive, observational, and cross-sectional study. Structured and semi-structured interviews were used to collect ethnopharmacological data. The plants used traditionally by the study population were collected and identified. An experimental study was performed to assess the giardicidal activity in vitro of the extracts from the selected plants.

Study population

Data were collected from patients aged over 18 years and caregivers of children treated at a municipal public primary healthcare facility (Unit A) and private institution specializing in digestive system diseases (Unit B), both facilities located in the city of Sao Luis, Maranhão state, Brazil.

São Luís, the capital of Maranhão state, Brazil, located at 2° 33' 00" S and 44° 18' 00" 19' W, has an area of 831.7 km², an estimated 1,053,922 inhabitants, and population density of 1215.69 inhabitants/km² (IBGE, 2013).

On the basis of a previous ethnopharmacological giardiasis survey in São Luis, Maranhão, Brazil (Amaral, 2007), considering 30% prevalence of plant use, 5% error, and 90% confidence interval, we surveyed 398 patients and/or caregivers as the study subjects.

Collection and analysis of ethnopharmacological data

To collect ethnopharmacological data, we used structured and semi-structured interviews with open and closed questions. Interviews were conducted from January to March 2011, and focused on the plant species used to treat diarrhea and dysentery.

This approach was chosen since people find it difficult to recognize giardiasis, but they can easily identify the plants used to treat diarrhea, the predominant giardiasis symptom that occurs in 90% of symptomatic patients (Lebwohl et al., 2003; Al-Mekhlafi et al., 2005). Dysentery was the disorder used in the above approach because it was difficult for interviewees to differentiate diarrhea from dysentery.

The respondents that mentioned the use of plants were also questioned about the form(s) of preparation, plant part(s) used, origin of the plant material, source of information, knowledge about possible side effects and contraindications, as well as their socio-economic characteristics.

Collection and botanical identification

The species cited in the ethnopharmacological survey were collected from areas mentioned by the respondents, including urban landscapes (parks, squares, backyards, and vegetable

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