Revista Brasileira de Farmacognosia 26 (2016) 379–384

Brazilian Journal of Pharmacognosy
REVISTA BRASILEIRA DE FARMACOGNOSIA
www.elsevier.com/locate/bjp

Original Article

Exudates used as medicine by the "caboclos river-dwellers" of the Unini River, AM, Brazil – classification based in their chemical composition

João Henrique G. Lago[a,b], Jaqueline Tezoto[a], Priscila B. Yazbek[a], Fernando Cassas[a], Juliana de F.L. Santos[c], Eliana Rodrigues[a,*]

[a] Department of Biological Sciences, Centro de Estudos Etnobotânicos e Etnofarmacológicos, Universidade Federal de São Paulo, Diadema, SP, Brazil
[b] Department of Exact Sciences and Earth, Universidade Federal de São Paulo, Diadema, SP, Brazil
[c] Coordenação em Ciência e Tecnologia, Universidade Federal do Maranhão, São Luís, MA, Brazil

ARTICLE INFO

Article history:
Received 30 June 2015
Accepted 14 March 2016
Available online 28 March 2016

Keywords:
Amazon forest
Ethnobotany
Medicinal plants
Ethnopharmacology
Exudates phytochemistry

ABSTRACT

Although the use of exudates in traditional medicine has been commonly observed during ethnopharmacological surveys, few records have been made concerning the scientific merits of these products. The aim of this study was to document ethnopharmacological data and to classify exudates used as medicine by the "caboclos" river-dwellers from the Unini River of Amazonas, Brazil, on chemical analyses basis. Using an ethnographic approach, indicated plants and their respective exudates were collected, identified and incorporated into herbarium of the National Institute of Amazonian Research. To classify these exudates, plant material was extracted using methanol, and obtained extracts were analyzed by Nuclear Magnetic Resonance and mass spectrometry aiming identification of main compounds. Fifteen exudates were indicated by "caboclos" river-dwellers as home remedies; among their therapeutic uses, inflammatory processes, culture-bound syndromes and respiratory diseases are most prominent. Based on their solubility and chemicals classes, fifteen exudates were classified into: latex (7), resins (5), sap (1), gum (1), oleoresin (1); and eleven of them have not been mentioned on pharmacological literature until this moment. The obtained results may contribute to chemical/pharmacological application of exudates from these species, several of which have been classically used in Brazilian folk medicine.

Introduction

Although the use of exudates in traditional medicine has been commonly observed during ethnopharmacological surveys, few records have been made concerning the scientific merits of these products. In Brazil, the use of resin has been noted among the ethnic groups of Kapoor (Balée, 1994), Paumari (Prance et al., 1987) and Waimiri Atroari (Milliken et al., 1992). A possible explanation for the gap in records is the difficulty in identifying the exudates, since these exudates resemble one another in physical appearance. However, their characterization and means of distinction have been facilitated in recent years, as a result of technological developments in chemistry, molecular biology and microscopy. Water-soluble exudates include gums, which are composed of polysaccharides and are secreted by wells; and saps, which consist of polysaccharides and amino acids, as described by Langenheim (2003). According to the same author, fat-soluble exudates include resins and oleoresins, which are composed of terpenoids and phenolic compounds secreted by resiniferous channels, cavities, trichomes and epidermal cells; oils are compounds of fatty acids and glycerol; and latex is a complex mixture of terpenoids, phenolic compounds, proteins and carbohydrates and it is secreted by laticifers.

Not only exudates are poorly studied from the ethnopharmacological standpoint, but, to the best of our knowledge, few pharmacological and chemical studies have been conducted on exudates. On the other hand, there are Amazonian cultures whose medicinal uses of exudates need to be identified and documented before they are lost due to the rapid introduction of synthetic medicines. Therefore, the aim of this study is to document ethnopharmacological data and classify, on the basis of chemical analysis, the exudates (as latex, resin, oleoresin and so on) used as medicine by "caboclos" river-dwellers from the Unini River of Amazonas, Brazil.

* Corresponding author.
E-mail: e.rodrigues@unifesp.br (E. Rodrigues).

http://dx.doi.org/10.1016/j.bjp.2016.03.001

Materials and methods

Ethnopharmacological data

The exudates analyzed in this study were indicated by twelve healers of "caboclos" river-dwellers living in two communities along the Unini River of Amazon, Brazil (Fig. 1), during 11 months of fieldwork, between April 2008 and January 2012 by one of the authors (JFLS). For local healing experts selection, a snowball sampling, as described by Bernard (1988), was performed by consultation of local inhabitants of riverine communities. Ethnographic techniques and methods were applied, including participant observation, field diaries, informal and unstructured interviews (Bernard, 1988; Foote-Whyte, 1990). During interviews, the following data sheets were administered: interviewee personal information, ethnopharmacological survey (ingredients, uses, parts used, mode of preparation, and contraindications of plants and animals used for therapeutic purposes) and plant collection (popular name, habit, time of flowering/fruiting, organoleptic and morphological aspects) (Santos et al., 2012). The exudates indicated by interviewees and their respective vegetable materials were collected during fieldwork at Terra Nova (01°41′ S, 61°49′ W) and Tapiira (01°46′ S, 62°13′ W) communities (Fig. 1), they were identified by Mr. José Ramos, plant taxonomy technician, and incorporated into the herbarium of the National Institute of Amazonian Research. Prior to the field work, all necessary permits were obtained for the study, including a permit access to the Conservation Units, in order to ensure collection of samples, transport of biological material and access to associated traditional knowledge, and prior informed consent of informants (SISBIO No. 16805-2, CGEN/MMA No. 47/2009 and CEP-UNIFESP/EPM, No. 1354/08).

Exudates extraction

Liquid exudates (latex, oleoresin and sap), were obtained from trunks of their respective trees by incisions utilizing machete or knife, depending on the thickness of the barks. A pointed metal cannula was adapted in the trunk of each tree for collection of exudates. The solids and semi-solids exudates (resin and gum, respectively), were stuck on tree trunks; therefore they were collected manually. All exudates were placed in amber glasses, properly labeled with date and numeric codes correlating them to the trees from which they were collected. These glasses were kept in a styrofoam box until they reach the laboratory where their extracts were prepared for chemical analysis. All exudates were dried after collection, before the preparation of the extracts.

Preparation of extracts and chemical analysis

Solubility of obtained exudates was evaluated using 10 mg of each plant material and 1 ml of H_2O or hexanes in a test-tube. Then, exudates were extracted using MeOH (50 ml of solvent to each 10 g of plant material) in a sonicated bath at room temperature during 20 min. After solvent evaporation under reduced pressure, obtained crude extracts were analyzed by ^{1}H and ^{13}C NMR (nuclear magnetic resonance) spectroscopy in a Bruker Avance 300 spectrometer (300 MHz to ^{1}H and 75 MHz to ^{13}C nuclei, respectively) using $CDCl_3$, CD_3OD, $(CD_3)_2SO$, $(CD_3)_2O$ or D_2O as solvents and internal standard. Crude extracts were also analyzed by HPLC/ESIMS (liquid chromatography/electrospray ionization mass spectrometry) using a Bruker Daltonics equipment Esquire 3000 Plus. HPLC system was coupled with a Zorbax-C18 (250 mm × 4.6 mm, 3.55 m, Agilent, USA) column at 40 °C. Solvents H_2O and acetonitrile (CH_3CN) were used, starting with 15% of CH_3CN (0–20 min), increasing to 100% (20–25 min), isocratic (5 min), and decreasing to 15% (30–32 min), and isocratic (3 min) at flow rate of 1 ml/min. Injection volume was 3 μl and UV detection at 352 nm and 280 nm. ESIMS spectra (low resolution) were recorded in full scan and product ion scan modes (argon CID). Ion source was set as follows: nebulizer gas = 3 l/min, desolvation gas = 15 l/min, DL = 150 °C, heat block = 300 °C and voltage = 3.5 kV. GC/EIMS (gas chromatography/low resolution electronic impact mass spectrometry) were measured in a Shimadzu GC-17A chromatograph equipped with a capillary column DB-5 (cross linked 5% phenyl in 95% silicone - 30 m, 0.32 mm, I.D., 0.25 μm film thickness) interfaced with a MS-QP-5050A mass spectrometer. Temperature programming was performed from 150 to

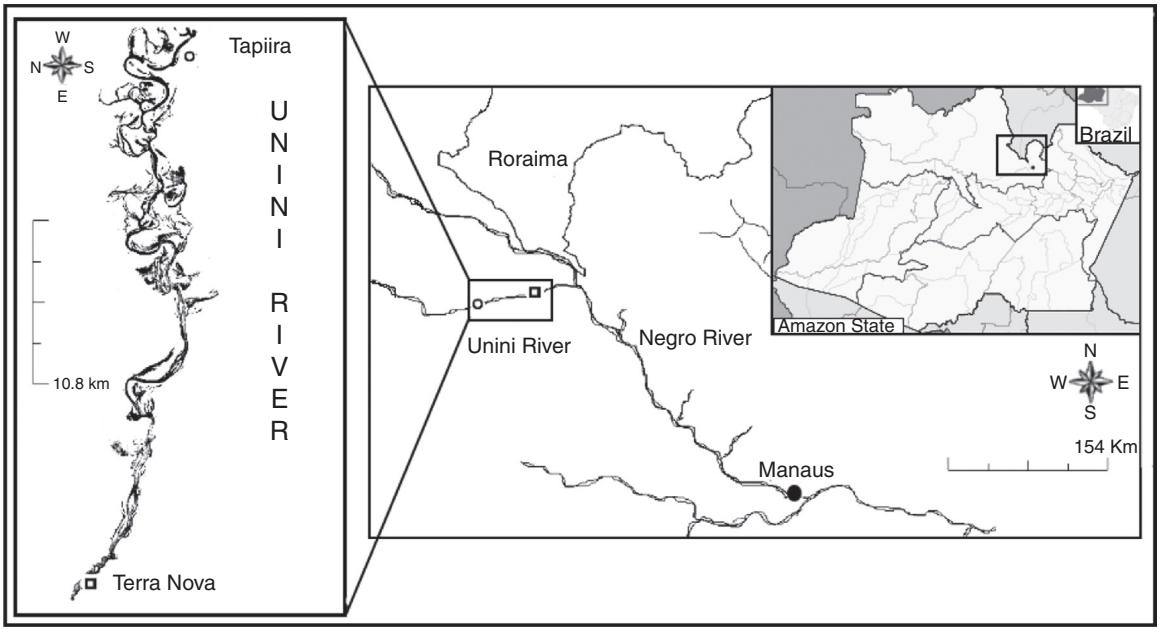

Figure 1. Unini River location in the Amazon forest biome, Amazon State – Brazil (map on the right). Location of two Unini River communities, where the exudates were collected during fieldwork: Terra Nova – 01°41′ S, 61°49′ W and Tapiira – 01°46′ S, 62°13′ W (left).
Source: Vitoria Amazônica Foundation (2005).