HOSTED BY
ELSEVIER

Contents lists available at ScienceDirect

Asian Pacific Journal of Tropical Medicine

journal homepage: http://ees.elsevier.com/apjtm

Original research http://dx.doi.org/10.1016/j.apjtm.2015.05.014

Brazilin from *Caesalpinia sappan* heartwood and its pharmacological activities: A review

Nilesh P. Nirmal[1*], Mithun S. Rajput[2], Rangabhatla G.S.V. Prasad[3], Mehraj Ahmad[4]

[1]*Center for Nutrition and Food Sciences, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Health and Food Sciences Precinct, Block 10, 39 Kessels Road, Coopers Plains, Acherfield Brisbane, QLD 4108, Australia*

[2]*Department of Medical Biochemistry, MGM Medical College, A.B. Road, Indore 452001, MP, India*

[3]*Biomedical and Pharmaceutical Technology Research Group, Nano Research for Advanced Materials, Bangalore 5600040, Karnataka, India*

[4]*Institute of Nutrition, Mahidol University, 999 Phutthamonthon 4 Rd., Salaya, Nakhon Pathom 73170, Thailand*

ARTICLE INFO

Article history:
Received 15 Mar 2015
Received in revised form 20 Apr 2015
Accepted 15 May 2015
Available online 25 June 2015

Keywords:
Brazilin
Caesalpinia sappan
Extraction
Heartwood
Pharamcological activities

ABSTRACT

Caesalpinia sappan L. (CS) is a plant of Leguminosae family, commonly known as Brazil or Sappan wood. CS is distributed in Southeast Asia and its dried heartwood has been used as traditional ingredient of food or beverages and has a wide variety of medicinal properties. Higher extraction yield of CS wood was achieved with 95% ethanol for 2 h. Chemical constituent's investigation of sappan wood resulted in the isolation of various structural types of phenolic components including one xanthone, one coumarin, three chalcones, two flavones three homoisoflavonoids and brazilin. Brazilin [(6a *S-cis*)-7, 11b-dihydrobenz[*b*]indeno[1,2-d]pyran-3,6a,9,10(6*H*)- tetrol], a major and active compound found in CS heartwood. Most of the folkloric uses of brazilin were validated by the scientific studies such as antioxidant, antibacterial, *anti*-inflammatory, *anti*-photoaging, hypoglycemic, vasorelaxant, hepatoprotective and *anti*-acne activity. CS heartwood extract is safe and did not produce any acute or subacute toxicity in both male and female rats. Brazilin is the safe natural compound having potential to develop as a medicinal compound with application in food, beverage, cosmetics and pharmaceutical industries to screen its clinical use in modern medicine. The information gained could provide the important and potential approach for pharmaceutical researcher to implicate the knowledge of brazilin in the formulation of new drug and to reveal therapeutic and gaps requiring future research opportunities. More studies are needed to evaluate the potential application of brazilin as preservative and coloring agent in food processing industries.

1. Introduction

Caesalpinia sappan L. is a plant of Leguminosae family, commonly known as Brazil or Sappan wood (Figure 1). *C. sappan* (CS) is distributed in Southeast Asia and its dried heartwood has been used as traditional ingredient of food or beverages [1]. The heartwood of the plant is commonly used for the extraction of red dye. The heartwood of CS has long been used in Thai folk medicine to treat tuberculosis, diarrhea, dysentery, skin infections and anemia [2]. Chemical constituent's investigation of sappan wood resulted in the isolation of various structural types of phenolic components including xanthone, coumarin, chalcones, flavones, homoisoflavonoids, and brazilin *etc* (Figure 2). Brazilin is the major compound naturally occurring in the CS heartwood and is used as a red dye for histological staining [3]. In traditional Chinese medicine, brazilin is used for treatment of increased blood circulation, promotes menstruation and exhibit analgesic and *anti*-inflammatory potentials [4]. Brazilin have been reported to possess various biological activities including antibacterial [5], *anti*-inflammatory [6], *anti*-photoaging [7], hypoglycemic [8,9], vasorelaxant [10,11], *anti*-allergic [12], *anti*-acne [13,14], antioxidant [15] and nuclease activity [16].

Researchers are more interested in finding bioactive compounds from natural origin as drugs, owing to either high cost of synthetic drug or side effects of the synthetic molecules. Therefore, there is a continuous evaluation of plant resources for

*Corresponding author: Nilesh P. Nirmal, Center for Nutrition and Food Sciences, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Health and Food Sciences Precinct, Block 10, 39 Kessels Road, Coopers Plains, Acherfield Brisbane, QLD 4108, Australia.
E-mail: nirmalnp21@yahoo.co.in

Peer review under responsibility of Hainan Medical University.

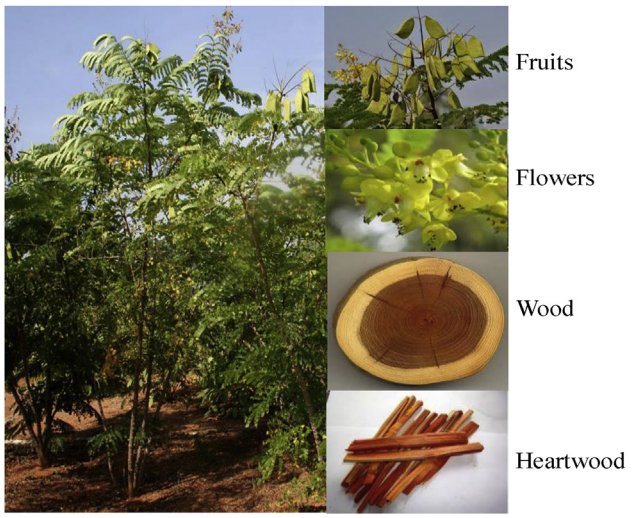

Figure 1. Photograph of *Caesalpinia sappan* plant with insight showing fruit, flower, wood and heartwood.

the search of bioactive molecule against non-medicated diseases. Brazilin is one of the most important bioactive natural compounds from CS heartwood having wide variety of industrial application including textile, food, beverage and pharmaceutical area. Since proven medicinal properties and used as dying agent, the wood has received both domestic and international market and being exported to USA and Europe from Southeast Asia [17]. Recently, CS heartwood has been listed in the 15th edition of the Japanese Pharmacopoeia [18]. In this review, we focus mainly on the different pharmacological activities of brazilin. Isolation techniques and structure of brazilin were also discussed. The information gained could provide the important and potential approach for pharmaceutical researcher to implicate the knowledge of brazilin in the formulation of new drug.

2. *Caesalpinia sappan* heartwood

The important part of CS is the heartwood which is pale red, hard, heavy with even and fine structure. The heartwood is traditionally used in Indian Ayurveda and Chinese folk medicine. In Thailand, it is mostly used as coloring agent in beverage, food, garment and cosmetics [19]. A decoction of heartwood is used in Namya-utai solution which has anti-thirst and cardiotonic properties. In Northern Thailand, especially in Chiang Mai, Nan and Lampang province, CS heartwood decoction is used as *anti*-inflammatory agent for the treatment of traumatic disease and arthritis [20]. The Northern Thai community has a long history of using decoction of CS heartwood for local consumption including health promotion and disease treatment. In Ayurveda, the heartwood is used for vitiated conditions of *pitta* which includes skin rashes, burning sensations, peptic ulcers, excessive body heat, heartburn and indigestion. It also used as blood purifier and in treatment of wounds, diarrhea, epilepsy, diabetes *etc.* CS heartwood is also used to reduce pain and swelling caused by external injuries and improvement of complexion [21]. In Chinese folk medicine, it is mainly used as an emmenagogue, hemostatic, analgesic and *anti*-inflammatory for the traumatic disease and blood flow promoting agent [22]. Additionally, decoction of CS heartwood is used for the treatment of blood pressure, burning sensations, cancer, cataract, digestion, dysmenorrhea, ear diseases, gonorrhea, heart diseases, jaundice, nervous disorders, obesity, ophthalmic diseases, spermatorrhoea, stomach aches, syphilis, urinary diseases and vascular diseases.

3. Brazilin

The heartwood contains water soluble flavonoids namely, brazilin, protosappanin and haematoxylin. Brazilin is the main homoisoflavonoid constituent found in the CS heartwood, which is well known as the natural red color dye for staining [3,16]. Brazilin also exhibit different industrial applications. Therefore, extraction and purification of brazilin is one of the important steps to achieve high extraction yield and purity, respectively.

4. Extraction and purification of brazilin

Extraction of phenolic compounds in plant materials is influenced by their chemical nature, the extraction method employed, sample particle size, storage time and conditions as well as the presence of interfering substances [23]. Therefore, extracts of plant materials are always mixture of different classes of phenolics that are soluble in the solvent system used. Solubility of phenolic compounds is governed by the type of solvent (polarity) used, degree of polymerization of phenolics, as well as interaction of phenolics with other food constituents and formation of insoluble complexes [24]. Therefore, there is no uniform or completely satisfactory procedure that is suitable for extraction of all phenolics or a specific class of phenolic substances in plant materials [25]. Methanol, ethanol, acetone, water, ethyl acetate and to a lesser extent propanol, dimethyl formamide and their combinations are frequently used for the extraction of phenolics. In some cases, mechanical means to enhance molecular interaction between the phenolic compounds and solvent are employed. Shaking, stirring, vortex mixing and sonication are four commonly used extraction techniques.

Traditional method for the extraction of red dye from sappan wood used boiling of wood pieces in water. During extraction few paddy grains are thrown into boiling solution to check the completion of red dye extraction. If the husk scales off, extraction is considered sufficient. In general, laboratory extraction from CS was carried out by using mainly ethanol (EtOH), methanol (MeOH) and water (H_2O) as solvents. Dried heartwood of sappan was extracted with MeOH for three days at room temperature [26]. The MeOH extract was concentrated and partitioned with H_2O–MeOH (3:1, 200 mL) and EtOAc (100 × 3). The EtOAc extract was concentrated and applied to silica gel column, eluted with $CHCl_3$–MeOH (15:1 to 5:1). The brazilin containing fraction was collected and loaded on sephadex LH-20 eluted with MeOH–H_2O (9:1) Brazilin was recrystallized from MeOH–H_2O [26]. CS heartwood powder (700 g) was extracted with methanol (2 L × 3, 1 h each) at 75 °C and extract was further concentrated to yield 60 g orange yellowish residue. The methanolic extract was dissolved in water and partitioned with hexane and ether. The ether fraction was further separated on silica gel column with $CHCl_3$–Me_2CO gradient system. Brazilin was purified to homogeneity with the help of preparative TLC [11]. Xu and Lee [5] extracted sappan heartwood powder with methanol by using reflux method. The methanol extract was dissolved in