PAU D'ARCO

Handroanthus spp. Mart ex DC Mattos

Family

Bignoniaceae (Jacaranda)

Parts Used

Dried inner bark.

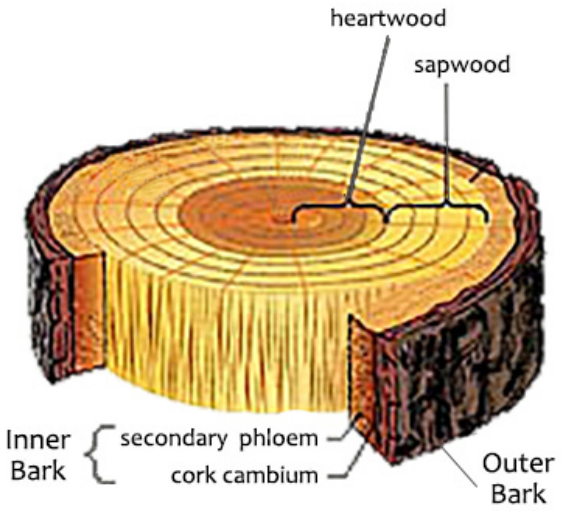
Description

Pau d'arco is an evergreen, canopy tree that can grow up to 38 metres high, and the base of the tree can be 2 to 3 metres in diameter. It is indigenous to the Amazon rainforest and the 'cerrado' (savannah) throughout Central and South America. Pau d'arco is Portugese for bow stick, which is an appropriate term considering the tree's use by the native South American Indians for making hunting bows. Its native names are ipê roxo (red thick bark), taheebo (ant wood), tajy ('to have strength and vigour' in Guarani and Tupi) or red (or purple) lapacho. In Australia its common name is trumpet tree.[1,2]

There is much confusion over the taxonomical division of the group of plants in the Bignoniaceae family, commonly and collectively known as pau d'arco, that have medicinal uses. The literature interchanges the genera *Tabebuia, Handroanthus* and *Tecoma*.

In relation to this, pau d'arco has been under scrutiny in regard to the correct identity of the species of plant matter used in fluid extract manufacture. It is quite possible that there is confusion among even trained gatherers. One specific way of distinguishing species is at the seedling stage. The four-leaf clover-like cotyledons are distinctly deeply cleft.[3]

Pau d'arco has recently undergone taxonomic revision and as a result the species name has changed. Formerly known botanically as *Tabebuia impetiginosa,* it has been reclassified as *Handroanthus impetiginosus* (Mart. ex DC.) Mattos. This is the accepted name and the former names are now synonyms.[4]



Synonyms: *Tabebuia impetiginosa* (Mart. ex DC.) Standley, *Tecoma impetiginosa* Mart. ex DC, *Tabebuia avellanedae* Lorentz ex Griseb, *Gelseminum avellanedae* (Lorentz ex Grisebach) Kuntze, *Handroanthus avellanedae* (Lorentz ex Griseback) Mattos, *Tabebuia palmeri* Rose, *Tecoma impetiginosa* var. *lepidota, Handroanthus impetiginosus* var. *lepidotus* Mattos, *Tecoma adenophylla* K. Schum ex Bureau and K.Schum, *Tecoma ipe* var. *integra* Sprague, *Tecoma integrum* (Sprague), *Tabebuia ipe* var. integra (Sprague) Sandwith, *Tecoma ipe* var. *integrifolia* Hassler, *Tecoma avellanedae* var. *alba* Lillo, T*abebuia nicaraguensis* S.F.Blake, *Tabebuia dugandii* Standley, *Tabebuia schunkevigoi* D.R.Simpson, *Tecoma avellanedae* (Lorentz ex Griseb.) Speg, *Tecoma impetiginosa* Mart., *Tecoma integra* (Sprague) Hassl.[5,6]

Over the course of its taxonomic history *Tabebuia* has been split and reassembled several times as researchers interpreted the morphological diversity in different ways. As they examined and monographed this taxon, a number of concepts of *Tabebuia* emerged, producing a labyrinthine synonymy (as demonstrated above).[7] The genus *Handroanthus* was erected by João Rodrigues de Mattos in 1970.[8] It was named for the Brazilian botanist Oswaldo Handro. *Anthus* is derived from a Greek word for flower. Most botanists at that time did not agree with the separation of *Handroanthus* from *Tabebuia*. *Tabebuia* is an abbreviation of "*tacyba bebuya*", a Tupi name meaning 'ant wood'.[9]

In 1992, a revision of the *Tabebuia* genus described 99 species and one hybrid.[10] In 2007 this large genus was revised again and on the basis of DNA, morphological and anatomical evidence subdivided into several genera. These consist of the 67 species and one hybrid that remain in *Tabebuia*, the two species transferred to *Roseodendron*, and the 30 species that are now placed in *Handroanthus*.[11]

Most species are characterised by the opposite, palmate (i.e. hand-like), compound leaves (a fully subdivided blade, each leaflet of the blade separated along a main, or secondary vein, leaf) with 3 to 5 leaflets, sometimes more, and large, showy, tubular, bell-shaped flowers (usually yellow or pink) and the elongated fruits containing numerous seeds. *Handroanthus* spp. includes species with long, narrow fruits rarely less than 20cm long and branched hairs are present at least on the midrib on the lower surface of the leaf. Flowers are yellow or pink usually with a prominent yellow throat.[12] *Handroanthus* spp. are also characterised by having dense wood, among the hardest and heaviest known, that is durable and attractive with extreme resistance to insects and fungal growth. The heartwood is distinct from the sapwood and contains copious quantities of lapachol.[13]

Tabebuia spp. includes those species with narrow fruits usually less than 15 cm long, and only scales and simple hairs are present on the lower surface of the leaf and branched hairs are lacking. Flowers are chiefly pink or white although *T. aurea* is yellow.[14] Pau d'arco lumber is in high demand in South America. The inner bark shavings commonly exported to the U.S.A and Europe are actually by-products of the South American timber and lumber industries. Prized for its dense, rot-resistant wood pau d'arco is among the most valuable Amazonian timbers, and is widely used for the construction of houses, boats and farm tools.[15]

Traditional Use

The inner bark of pau d'arco has been used as a cure-all by the native Americans of Brazil, northern Argentina, Paraguay, Bolivia and Peru for thousands of years.Traditionally it is ingested as a decoction, with indications that may pre-date the Incas (13th Century), as an analgesic, anti-inflammatory, astringent, antineoplastic and diuretic for treating inflammatory diseases, lupus, cancer, diabetes, prostatitis, blood coagulation, fungal, bacterial and viral infections including syphilis, candidiasis, malaria, fevers, trypanosomiasis (the name of several diseases in vertebrates caused by parasitic protozoan trypanosomes of the genus Trypanosoma), and gastrointestinal problems including constipation and peptic ulcers.[16,17,18,19,20,21,22,23]

As early as 1873, Dr. Joaquin Almeida Pinto had already described many of the therapeutic properties of pau d'arco. He prescribed it as a fever reducer and the bark was used against ulcers. He also used it for venereal, rheumatic and skin disorders, especially eczema, herpes and the mange.[24]

Pau d'arco has been acclaimed as one of the 'miraculous' cures for cancer and tumours. It was during the 1960s that it attracted considerable attention in Brazil and Argentina as a 'wonder drug'. In 1967, after reports in the Brazilian press, it came back into fashion with clinicians and the public in general. The news magazine O'Cruzeiro (March 18 and March 25, 1967) started reporting 'miraculous' cures in cancer patients in the hospital clinics of Sao Paulo and in the Municipal Hospital of Santo Andre.[25,26]

The O'Cruzeiro reporters tracked down the people who were championing the bark: Dr. Valter Accorsi, a botanist at the University of Sao Paulo and Dr. Orlando dei Santi. These reports highlighted the miraculous curing of a seriously ill, young patient. Dr. dei Santi came into contact with pau d'arco during a dinner in Sao Paulo hosted by a family returning from Rio de Jainero. During the dinner Dr. dei Santi heard the story of a young girl from Rio that, while gravely sick with cancer, had been given up on by the medical establishment. A great aunt of the girl had contacted a tribal medicine man who had told her that the girl could be cured using the bark of pau d'arco.

The family of the girl would not follow the advice at first but after the girl had not one, but two, strange dreams where a monk promised her recovery should she drink the tea brewed from the bark of pau d'arco trees growing in Pernambuco and Bahía (northeast Brazil), the family heeded the advice and the girl regained her health. The most immediate result was the cessation of all pain followed by the complete disappearance of the malignancy. After hearing this account Dr. dei Santi returned to the hospital in Santo Andre where his brother was sick with cancer, laying near death. Contrary to what he was taught in medical school against the use of empirical remedies used by Indian tribes, Dr. dei Santi decided to test a decoction of pau d'arco on his dying brother. It was reported that he concocted a brew of dry pau d'arco bark in white wine, mixed with orange juice, and administered it to his brother while fasting.[27] After a month of uninterrupted treatment with the concoction Dr. dei Santi's brother was discharged from hospital without a trace of cancer. After this Dr. dei Santi started to treat his patients with pau d'arco and the practice spread rapidly among other physicians in the hospital who used the unapproved medicine for viral-linked diseases including leukaemia.[28,29,30]

Dr Accorsi was contacted by O'Cruzeiro reporters while travelling to Piracicaba.31 There he was treating around 2000 people a day with tea made from pau d'arco bark. After hearing about the case of the girl in Rio, Dr Accorsi started studying pau d'arco trees in his state in Sao Paulo and comparing them with the trees in Bahia and Pernambuco. By comparing the effects in leukaemia patients he realized that the Bahia trees had a more potent effect in patients and was able to verify 'two great truths': first, that pau d'arco tea eliminated the pain caused by the disease and second that it also caused a significant increase in red blood cells.[32,33,34]

Independent of Dr. dei Santi and Dr Accorsi, Dr. Theodore Meyer, Professor of Botany and Plant Geography at the National University of Tucuman in Argentina, documented the first clinical cases of cancer chemotherapy using extracts from various pau d'arco inner barks in 1966. Dr. Meyer's work, though it lacked adequate controls and statistical evaluations, provided observational evidence for the efficacy of pau d'arco but he was not able to convince the medical community of his findings before dying in 1972.[35,36]

Constituents

The major active constituents are 16 quinones, containing both naphthoquinones: (lapachol 2-7% [2-hydroxy-3-(3-methyl-2-butenyl)-1,4-naphtalene-dione], menaquinone-I, deoxylapachol, beta-lapachone, alpha-lapachone and dehydro-alpha-lapachone) and anthraquinones: (2-methylanthraquinone, 2 hydroxymenthylanthraquinone, 2-acetoxymethylanthraquinone, anthraquinone-2-aldehyde, 1-hydroxyanthraquinone, 1-methoxyanthraquinone, 2-hydroxy-3-methylquinone and tabebulin). It is considered very rare to have both these groups of quinones occurring in the same plant.[37] Several of the remarkable properties of pau d'arco may be due to a probable synergy between naphthoquinones and anthraquinones.[38] Other constituents include tabebuin, lapachenole, quercetin, o- and p-hydroxybenzoic acids, benzaldehyde derivatives, cyclopentene dialdehyde, flavonoids, iridoid glycosides, lignan glycosides, isocoumarin glycosides, phenylethanoid glycosides and phenolic glycosides.[39,40,41,42]

Actions

Antitumour, antioxidant, antimicrobial, antiviral, antiparasitic, anti-inflammatory, anticoagulant, antiplatelet; analgesic; antiproliferative, antidepressant, antiobesity.

Pharmacological Activity

To date there are no well-defined controlled clinical trials in humans to support pau d'arco as a monotherapy for any condition. Due to this, much of the information is based on traditional use or evidence of activity.

Lapachol was originally identified as the 'signature' compound in pau d'arco and much of the earlier pharmacological research was focused on it. More recently, its significance has been questioned. In particular, beta-lapachone possesses marked antimicrobial and antitumour activities and has been the main focus of current research.[43] There are studies which show significantly better results with the whole extract and diminishing effectiveness as the extracts are refined or individual chemicals are tested.[44]

Most of the chemical analyses of pau d'arco have been performed on the heartwood of the tree, rather than on the phloem, or inner lining of the bark, which is used medicinally. Traditionally it is the living bark of a plant, especially a tree or shrub, that is used medicinally and not the heartwood. The reason is simple: the nutrients and representative families of chemical substances used to sustain the life of the tree are found in greatest concentration in the cambium layer and phloem of the living bark. The outer bark and heartwood are, essentially, inactive materials that only serve to provide strength to the tree.[45] This may explain why a 1994 review of pau d'arco products on the Canadian market found no or low levels of lapachol in all of the products. While poor quality of products is probably one reason for this finding, levels of lapachol also depend on whether the inner bark or wood is used. Typical levels of total naphthoquinones expressed as 'lapachol' are 1 to 2% for the inner bark, but lapachol itself is likely to be only a minor constituent of this plant part.[46]

The rationale of using the inner bark of pau d'arco is exquisitely explained in popular terms by Mowrey:

'The life processes of a mature tree are carried out in the thin corridor lying between the outer bark and the inner heartwood. Pull the bark off a tree and you will notice moist, very thin layers of tissue that seem to shred when picked at with the hands. This is the cambium layer. Its purpose is to create new tree tissues, such as phloem, through cell division. The newest, youngest phloem cells are just outside the cambium. As new phloem is added, older cells are crushed and pressed into the bark. Younger, newer cells added to the inside of the cambium layer are called xylem. Newer xylem is called sapwood; older xylem is crushed and pressed into the heart of the tree. It is therefore known as heartwood. The actively conducting tissues of a tree are the thin layers of fresh xylem and phloem on each side of the cambium. Indiscriminate combining of older, less active layers of bark and tree with the younger, living tissues results in a dramatic dilution of active principle and medicinal value. Yet it is a common practice.' [47]

Antitumour Activity

Pau d'arco constituents have shown activity against the following cancer cell lines: carcinoma (Walker

256), prostate cancer (DU-145, PC-3, LNCaP),[48] human promyelocytic leukaemia (HL-60), breast carcinoma, ovarian carcinoma, epidermoid laryngeal carcinoma (HEp-2),[49] radio-resistant human malignant melanoma (U1-Mel),[50] human breast cancer (MCF-7:WS8),[51] human lung adenocarcinoma (A549), human colon cancer cells, human cervical cancer (HeLa) and osteosarcoma (HuO9).[52,53,54]

Pau d'arco showed enhanced antiproliferative activity against lung cancer cell lines in a study evaluating the antiproliferative activity, antioxidant capacity and tannin content in 14 species of plants from semi-arid northeastern Brazil (Caatinga). The methanol extracts were assayed for antiproliferative activity against the HEp-2 (laryngeal cancer) and NCI-H292 (lung cancer) cell lines using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazole method. It was suggested that future *in vitro* and *in vivo* comparative studies with other pharmacological models be conducted, and that a process of purification and identification of the possible molecule(s) responsible for the observed pharmacological activity should be started.[55]

Both pau d'arco (30 to 500mg/kg) and the napthoquinone, beta-lapachone (1 to 5mg/kg), equally prolonged the life span in Ehrlich ascites tumour-bearing mice by reversing myelosuppression (bone marrow suppression), which is the decrease in cells responsible for providing immunity, carrying oxygen, those responsible for normal blood clotting, and which can be a serious side effect of chemotherapy especially for leukaemia. The myelosuppression was concomitant with increases in spleen granulocyte and macrophage progenitor cells, and in serum colony-stimulating activity, and these effects were reversed in a dose-dependent manner. The optimal biologically active doses of 120mg/kg pau d'arco and 1mg/kg beta-lapachone prolonged the life span of tumour-bearing mice, both producing the same rate of extension in the duration of survival. Toxic manifestations were produced by the higher doses of beta-lapachone in normal and tumour-bearing mice. In spite of similarities between treatments, pau d'arco concentrations used to treat the animals presented no traces of beta-lapachone, as measured by TLC and HPLC analyses. The findings suggest that the antitumour effect of pau d'arco and beta-lapachone, acting synergistically with other factors such as specific cytokines, may result from enhanced macrophage activation against tumour cells.[56]

A stereoisomer (two molecules made of the same atoms, connected in the same sequence, but the atoms are positioned differently in space) of a biologically active naphthoquinone from pau d'arco has been shown to exhibit potent cytotoxicity against several human tumor cell lines, while displaying lower cytotoxicity than mitomycin (a chemotherapeutic agent) against some human normal cell lines. The enantiomer (one of two stereoisomers that are mirror images of each other that are not identical) of this compound was less active against tumor cell lines.[57] A synthetic version of the natural product beta-lapachone has also demonstrated promising anticancer activity. ARQ 501 formulated with hydroxypropyl-beta-cyclodextrin has successfully completed phase I clinical trials and is currently in several phase II human clinical trials for the treatment of pancreatic cancer, head and neck cancer, and leiomyosarcoma (smooth muscle connective tissue tumour).[58]

A 2013 study has highlighted the importance of furonaphthoquinones as antileukemic agents. Furonaphthoquinones are promising skeletons for anticancer drug molecules. In particular, methoxylated furonaphthoquinones are characteristic constituents of pau d'arco plants. The study demonstrated that the synthetic furonaphthoquinones showed moderate cytotoxicity against human leukemia U937 and HL-60 cells.[59]

For thirty years, between 1960 and 1990, the National Cancer Institute in the U.S.A (NCI) carried on a program for the extensive recollection and screening of plants in temperate regions, with the aim of isolating compounds effective in cancer chemotherapy.[60] One of the compounds isolated as part of this effort, lapachol, was identified as the compound responsible for the anticancer activity of pau d'arco.[61] Lapachol entered phase I clinical trials at the NCI in 1968, on the basis of its activity against Walker 256 tumours (with a confidence rate exceeding 90%). However, as soon as effective plasma levels were attained undesirable side effects were severe enough to require that the drug be stopped. These included moderate to severe nausea, vomiting and antivitamin K activity (anaemia and a tendency to bleed). Interestingly,

other chemicals in the whole plant extract (which, initially, showed positive antitumor effects and very low toxicity) demonstrated positive effects on vitamin K and compensated for lapachol's negative effect. Instead of pursuing research on a complex combination of at least 20 active chemicals in a whole plant extract (several of which had antitumor effects and other positive biological activities), research focused on a single, patentable chemical and it didn't work as well. The investigative new drug (IND) status for the drug was withdrawn in 1970.[62,63]

Despite NCI's abandonment of the research, another group developed a lapachol analog (which was patentable) in 1975. A dichloroallyl lawsone (DCL), a synthetic analogue of the antimalarial lapachol, which had a better *in vivo* activity in the Walker 256 system, was selected to replace lapachol with IND approval in 1975. Like lapachol, DCL was found to be an inhibitor of oxidative phosphorylation (the metabolic pathway in which the mitochondria in cells use their structure, enzymes and energy released by the oxidation of nutrients to reform ATP [the molecule that supplies energy to metabolism]). In Rhesus monkeys the agent was observed to have some cardiac toxicity, subsequently it was decided that there was little point to any further analogue development and the case was closed.[64] One study reported that this lapachol analog increased the life span of mice inoculated with leukemic cells by over 80%.[65]

Antioxodant Activity

The antioxidant activity of the volatile constituents of pau d'arco dried inner bark were evaluated using two different assays and was comparable with that of the well-known antioxidants, alpha-tocopherol and butylated hydroxytoluene (a common food additive). The extract exhibited a potent inhibitory effect on the formation of conjugated diene hydroperoxides (from methyl linoleate) at a concentration of 1000 micrograms/mL. The extract also inhibited the oxidation of hexanal for 40 days at a level of 5 micrograms/mL.[66,67]

Antimicrobial Activity

As early as 1956, pau d'arco clearly demonstrated broad clinical actions against a number of disease-causing microorganisms supporting its wide array of uses in herbal medicine. Antimicrobial properties of many of pau d'arco's active phytochemicals were demonstrated in several clinical studies in which they exhibited strong *in vitro* activity against various gram-positive and gram-negative bacteria, fungi, and yeast (including *Candida albicans*, *Aspergillus*, *Staphylococcus*, *Streptococcus*, *Helicobacter pylori*, *Brucella*, tuberculosis, pneumonia, and dysentery.)[68,69,70,71,72] In addition to its isolated chemicals, a hot water extract of pau d'arco demonstrated antibacterial actions against *Staphylococcus aureus*,[73] *Helicobacter pylori* (the bacteria that commonly causes stomach ulcers)[74] and *Brucella*.

Lapachol, like many napthoquinones, acts as a respiratory poison by interfering with electron transport in microbes. This mechanism was first demonstrated on malarial parasites by inhibition of oxygen uptake.[75] At 100mg/L concentration, lapachol has shown to inhibit the uptake of oxygen in *Plasmodium knowlesi* (a primate malaria parasite) by 74% and the succinate oxidase system by 26%. These results lead to the conclusion that lapachol exhibits antimalarial activity against *Plasmodium knowlesi* via respiratory inhibition as a likely mechanism.[76,77]

Compounds identified in pau d'arco inner bark have been shown in an *in vitro* study to inhibit pathogenic bacteria in the human intestinal tract while having no adverse effect on beneficial probiotic strains. Anthraquinone-2-carboxylic acid and lapachol were evaluated for their growth-inhibiting activity toward 10 human intestinal bacteria using a paper disk diffusion bioassay (a test which uses antibiotic-impregnated wafers to test whether partcular bacteria are susceptible to specific antibiotics) and compared to those of seven lapachol congeners (1,4-naphthoquinone, naphthazarin, menadione, lawsone, plumbagin, juglone, and dichlone) as well as two commercially available antibiotics, chloramphenicol and tetracycline. Anthraquinone-2-carboxylic acid exhibited very strong growth inhibition of *Clostridium paraputrificum* at 1 microg/disk while 100 microg/disk of lapachol was needed for moderate growth inhibition of the same organism. These two isolates exhibited weak inhibition of *Clostridium perfringens* and *Escherichia coli* at 100 microg/disk while no adverse effects were observed on the growth of

Bifidobacterium adolescentis, *Bifidobacterium bifidum*, *Bifidobacterium infantis*, *Lactobacillus acidophilus* and *Lactobacillus casei* at 1000 microg/disk. Structure-activity relationships indicate that a methyl group in the C-2 position of 1,4-naphthoquinone derivatives might play an important role in antibacterial activity.[78]

An *in vitro* study showed that pau d'arco contains compounds with anti-*Helicobacter pylori* activity. The biologically active components of pau d'arco dried inner bark were characterized by spectroscopic analysis as 2-(hydroxymethyl) anthraquinone, anthraquinone-2-carboxylic acid, and 2-hydroxy-3-(3-methyl-2-butenyl)-1,4-naphthoquinone (lapachol). The growth-inhibiting activity of these constituents against *Helicobacter pylori* were examined using paper disc diffusion and minimum inhibitory concentration (MIC) bioassays. The activity of the isolated constituents was compared to that of the commercially available anti-*Helicobacter pylori* agents, amoxicillin, metronidazole and tetracycline.[79]

An analogue of lapachol (furanonaphthoquinone) was shown to significantly lower MIC against methicillin-resistant *Staphylococcus aureus* (MRSA) strain compared to methicillin-sensitive *Staphylococcus aureus* (MSSA). The finding was statistically significant with a probability-value of <0.01 (there is a one percent chance that the result was accidental). After a major investigation of the pharmacological properties of the compounds isolated from pau d'arco inner bark, the quinones beta-lapachone, 3-hydroxy beta N lapachone and alpha-lapachone can be used successfully in topical preparations against wound infections caused by staphylococci. The quinones were tested to determine the MIC values against methicillin-resistant *Staphylococcus aureus*, *Staphylococcus epidermidis* and *Staphylococcus haemolyticus* strains, (the latter two hetero-resistant to vancomycin, the last therapy used in these cases). The quinones showed antibacterial activity (MICs of 8, 4/8 and 64/128 microg/mL to beta-lapachone, 3-hydroxy beta N lapachone and alpha-lapachone, respectively), but no bactericidal activity was observed (MBC > 512 microg/mL for all compounds).

Also it was found that lapachol has efficacy against *Helicobacter pylori*, *Staphylococcus*, *Streptococcus*, *Enterococcus*, *Bacillus* and Clostridium species with MIC ranging from 1.56 to 25mcg/mL. In addition it was reported that lapachol has a relevant effect against *Candida albicans*, *Candida tropicalis* and *cryptococcus neoformans* that was similar to Amphotericin B. The presumed antifungal activity of lapachol is believed to be due to its interaction with the cellular membrane.[80,81,82,83]

Antiviral Activity

The naphthoquinones of pau d'arco, in particular lapachol, have been stated to be active against certain viral strains including herpes virus types I and II, influenza, poliovirus, and vesicular stomatitis virus.[84] The mechanism of action of these quinones is proposed to be via deoxyribonucleic acid (DNA) and ribonucleic acid (RNA) polymerase inhibition and retrovirus reverses transcriptase. Furthermore β-lapachone is presumed to interfere with the replication of type 1 human immunodeficiency virus (HIV-1) via transcriptase inhibition. Transcription of HIV-1 provirus is governed by the viral long terminal repeat (LTR). Drugs can block HIV-1 replication by inhibiting activity of its LTR. Beta-lapachone is a potent and selective inhibitor of HIV-1 LTR-directed gene expression, at concentrations that have minor effects on cells.[85]

Antiparasitic Activity

The antiparasitic action of pau d'arco seems to come from increasing oxygen supply to the local level, destroying bacteria, viruses, fungi and parasites.[86] Pau d'arco has been used orally, and as a topical barrier, to be effective against skin penetration to trematodes (parasitic flatworms or flukes) specifically *Schistosoma mansion*, which causes the common tropical disease schistosomiasis. This parasite lives in water and enters the host by penetrating through the skin. It can cause a complicated disease which can sometimes be fatal. In addition, lapachol is claimed to have some effect against *Trypanosoma cruzi*, which causes trypanosomiasis or Chaga's disease.[87] Leishmania is a genus of trypanosomatid protozoan parasites responsible for the disease leishmaniasis. It is transmitted to humans by any of several species of sand flies. An amastigote is a cell that does not have a visible external flagella or cilia.

The term is used mainly to describe a certain phase in the life-cycle of trypanosome protozoans.

It is also called the leishmanial stage, since in Leishmania it is the form the parasite takes in the vertebrate host, but occurs in all trypanosome genera.[88] Pau d'arco has shown to induce lysis (breaking down of a cell) of intracellular amastigotes *in vitro*, exhibiting marked leishmanicidal activity.[89,90]

Anti-inflammatory and Analgesic Activity

A 2013 study has suggested that beta-lapachone may be effective in the treatment of inflammatory diseases such as multiple sclerosis (MS), an autoimmune disorder characterized by central nervous system (CNS) inflammation and demyelination. As mentioned above, beta-lapachone is currently being evaluated in clinical trials for the treatment of cancer but recent investigations also suggest its potential application for treatment of inflammatory diseases. Reactive T cells, including IL-17 and IFN-γ-secreting T cells, are believed to initiate MS and the associated animal model system experimental autoimmune encephalomyelitis (EAE). IL-12 family cytokines secreted by peripheral dendritic cells (DCs) and CNS microglia are capable of modulating T-cell phenotypes. The present studies demonstrated that beta-lapachone selectively inhibited the expression of IL-12 family cytokines, including IL-12 and IL-23 by DCs and microglia, and reduced IL-17 production by CD4(+) T-cells indirectly through suppressing IL-23 expression by microglia. Importantly, the studies also demonstrated that beta-lapachone ameliorated the development on EAE. Beta-lapachone suppression of EAE was associated with decreased expression of mRNAs encoding IL-12 family cytokines, IL-23R and IL-17RA, and molecules important in Toll-like receptor signalling. Collectively, these studies suggest mechanisms by which beta-lapachone suppresses EAE and suggest that beta-lapachone may be effective in the treatment of inflammatory diseases such as MS.[91]

A 2013 rodent study evaluated the gastric ulcer healing property of the ethanolic extract (EET) of bark from pau d'arco and investigated the mechanisms that may underlie its anti-inflammatory effect. Rats were treated with EET (twice a day for 7 days) after induction of chronic gastric ulcers by 80% acetic acid.

Following treatment, histological and immunohistochemical analysis were performed in gastric ulcer tissues. Oral administration of EET (100 and 300 mg/kg) significantly reduced the gastric lesion induced by acetic acid in 44 and 36%, respectively. Histopathological evaluation demonstrated a contraction of gastric ulcer size, increase of mucus layer and cell proliferation in animals treated with EET (100 and 300mg/kg). The results demonstrate that EET significantly accelerates healing of acetic acid induced gastric ulcer in rats through increase of mucus content and cell proliferation, indicating a potential usefulness for treatment of peptic ulcer diseases.[92]

The aqueous extract of pau d'arco significantly suppressed inflammatory mediators in various *in vitro* and *in vivo* inflammatory conditions, and therefore may be considered as a therapeutic remedy for various inflammatory diseases such as arthritis and atherosclerosis. Pau d'arco substantially inhibited the production of prostaglandin (PGE2) and nitric oxide (NO), and blocked the mRNA expression of their catalyzing enzymes (cyclooxygenase [COX)-II] and inducible NO synthase [iNOS], respectively), in lipopolysaccharide (LPS)-stimulated mouse leukemic monocyte macrophage cell line. The blockade of inflammatory mediators seemed to be the result of the interruption of extracellular signal-related kinase (ERK) activation, according to immunoblotting analysis and the NO assay, where LPS strongly induced the phosphorylation (a hallmark of activation) of ERK, and U0126, a selective ERK inhibitor, was found to strongly inhibit PGE(2) production. Similarly, oral administration of pau d'arco (100mg/kg) for one week completely diminished mouse ear oedema induced by arachidonic acid, an activator of COX-II, but not croton oil, an activator of lipoxygenase, suggesting that the ethnopharmacological action of pau d'arco may be due to its negative modulation of macrophage-mediated inflammatory responses by suppressing PGE2 production.[93]

In 2000 a study isolated two compounds from the bark of pau d'arco which showed anti-inflammatory activity. They are cyclopentene dialdehydes, 2-formyl-5-(4'-methoxybenzoyloxy)-3-methyl-2-cyclopentene-1-acetal dehyde, and 2-formyl-5-(3', 4'-dimethoxybenzoyloxy)-3-methyl-2-cyclopentene-1-acetaldehyde. The structures were established by analysis of spectroscopic data.[94]

The results of a 2012 study suggest that the ethanolic extract of the inner bark of pau d'arco has the potential to be developed as a therapeutic, or supportive drug, against diseases with accompanying pain and inflammation, including osteoarthritis. In the study, various animal models were used to demonstrate the analgesic and anti-inflammatory properties of an ethanolic extract of pau d'arco, thereby investigating its potential as a therapeutic treatment for diseases with pain and inflammation. In the hot plate and writhing tests for the *in vivo* analgesic effect test of pau d'arco, a 200mg/kg dose of the extract induced a significant antinociceptive effect and increased the pain threshold by approximately 30% compared with the control. In vascular permeability and tetradecanoylphorbol acetate (TPA)-, arachidonic acid- and carrageenan-induced paw oedema tests for anti-inflammatory effects, treatment with 200mg/kg pau d'arco led to significant anti-inflammatory effects and inhibited inflammation by 30-50% compared with the control. At 100mg/kg, the extract decreased the levels of pain and inflammation in all tested models, but the degree of inhibition was not statistically significant.[95]

An extract of pau d'arco inner bark demonstrated antinociceptive (reducing sensitivity to painful stimuli) and antioedema activities when used in various rat studies. At 200mg/kg, the aqueous extract of pau d'arco inhibited the rat paw oedema induced by carrageenan in a similar way as indomethacin. This test is used to evaluate anti-inflammatory drugs and has been used to test the antioedema effect of various substances. Furthermore, the inner bark aqueous extract, administered orally in three different concentrations, 100, 200 and 400mg/Kg, reduced the nociception produced by formalin acetic acid (0.6% in water, i.p.) by 49.9%, 63.7% and 43.8%, respectively. The formalin test is one of the most used models to explain pain and analgesia mechanisms, with better results than the ones using mechanical or thermal stimulus.[96,97,98]

An *in vitro* study revealed that lapachol and its analogues to have an antipsoriatic effect by inhibiting the growth of human keratinocyte cell line HaCaT, (a cell type belonging to an immortal human keratinocyte line used in scientific research) and reducing inflammation.

The authors of the study concluded that pau d'arco has similar antipsoriatic activity as anthralin in inducing damage to the cell membranes of the keratinocyte cells.[99]

Anticoagulant, Antiplatelet and Antiproliferative Activity

An *in vitro* study using washed rabbit platelets and cultured rat aortic vascular smooth muscle cells showed that various extract fractions of pau d'arco have antiplatelet and antiproliferative activities which may be of benefit in cardiovascular disease. The n-Hexane, chloroform and ethyl acetate fractions selectively inhibited platelet aggregation induced by collagen and arachidonic acid (AA) in a dose-dependent manner. Also, the chloroform fraction in particular, significantly suppressed AA liberation induced by collagen in rabbit platelets as well as potently inhibited cell proliferation and deoxyribonucleic acid (DNA) synthesis induced by platelet derived growth factor.[100]

Lapachol has been shown to be a potent inhibitor of both vitamin K epoxide and quinone reductase of rat liver microsomes *in vitro*. These observations explain the anticoagulant activity of lapachol previously observed in both rats and humans.[101]

Antidepressant Activity

Results of a 2013 study to investigate the ability of pau d'arco ethanolic extract to reverse depression in mice indicate that this plant could constitute an attractive strategy for the management of depressive disorders. The aim of the study was to investigate the ability of chronically administered pau d'arco to cause an antidepressant-like effect in the tail suspension test (TST), a predictive test of antidepressant activity, and to reverse behavioural (hyperactivity, anhedonic-like behaviour and increased immobility time in the TST) and biochemical changes induced by olfactory bulbectomy (OB), a model of depression, in mice. Pau d'arco ethanolic extract's ability to abolish the behavioural changes induced by OB was accompanied by modulation of ERK1 and BDNF signalling pathways, being a promising target of EET. Mice were submitted to OB to induce depressive-related behaviours, which were evaluated in the open-field test (hyperactivity), splash test (loss of motivational and self-care behaviour indicative of an anhedonic-like behaviour) and TST (increased immobility time).

Phosphorylation levels of Akt, GSK-3β, ERK1/2 and CREB, as well as BDNF immunocontent, were evaluated in the hippocampus of bulbectomized mice or sham-operated mice treated for 14 days by p.o. route with EET or vehicle. The results showed that EET (10 and 30mg/kg) given 14 days by p.o route to mice reduced the immobility time in the TST without altering locomotor activity, an indication of an antidepressant-like effect. EET per se increased both CREB (Ser(133)) and GSK-3β (Ser(9)) phosphorylation (at doses of 10 to 30 and 30mg/kg, respectively) in sham-operated mice. OB caused hyperactivity, loss of motivational and self-care behaviour, increased immobility time in the TST and an increase in CREB and ERK1 phosphorylation, as well as BDNF immunocontent. EET abolished all these OB-induced alterations except the increment of CREB phosphorylation. Akt (Ser(473)) and ERK2 phosphorylation levels were not altered in any group.[102]

The follow up 2013 study was aimed at investigating the contribution of N-methyl-D-aspartate (NMDA) receptors and the L-arginine-nitric oxide (NO)-cyclic guanosine 3'5'-monophosphate (cGMP) pathway to the antidepressant-like action of the ethanolic extract from pau d'arco in the tail suspension test (TST). The anti-immobility effect of the extract (30 mg/kg, orally [p.o.]) was prevented by pretreatment of mice with NMDA, L-arginine and sildenafil. Additionally, the combination of MK-801, 7-nitroindazole and 1H-[1,2,4]oxadiazole[4,3-a] quinoxalin-1-one (ODQ) with a subeffective dose of EET (1 mg/kg, p.o.) produced a synergistic antidepressant-like effect in the TST, without causing significant alterations in the locomotor activity. Moreover, the administration of an effective dose of EET (30mg/kg, p.o.) produced a reduction in NOx levels in the cerebral cortex. Conversely, a subeffective dose of EET (1mg/kg, p.o.) caused no changes in the cortical NOx levels. Results suggest that the antidepressant-like effect of EET in the TST is dependent on a blockade of NMDA receptor activation and inhibition of NO-cGMP synthesis, significantly extending literature data about the antidepressant-like action of this plant and reinforcing the notion that this plant may be useful in the management of depressive disorders.[103]

Antiobsesity Activity

The results of a 2014 study suggest that an ethanolic pau d'arco extract inhibits obesity and fat accumulation by regulation of gene expression related to lipid metabolism in high-fat diet (HFD)-induced obesity in mice. Mice were fed a HFD (25% fat, w/w) for 11 weeks. Mice administered pau d'arco had significantly reduced body weight gain, fat accumulation in the liver, and fat pad weight, compared to HFD mice. Reduced hypertrophy of fat cells was also observed in pau d'arco mice. Mice administered pau d'arco also showed significantly lower serum levels of triglycerides, insulin and leptin.[104]

Indications

- Adjunct for Cancer
- Inflammatory diseases including lupus, psoriasis, cardiovascular disease
- Infections, candidiasis, trichomonas vaginalis, herpes simplex virus, influenza, *Helicobacter pylori*
- Parasitic diseases such as schistosomiasis
- Cervicitis and cervico-vaginitis
- Oedema

Energetics

Cold, astringent, bitter.

Use in Pregnancy

Contraindicated in women who are pregnant, or trying to become pregnant, due to fetotoxic and abortifacient effects of lapachol, based on animal studies. Caution is also warranted in the male partners of patients attempting to become pregnant as reproductive toxicity has been reported in animal studies.[105,106,107]

Contraindications

Caution should be used in patients with blood disorders, those who are having surgery, or those who are taking anticoagulant, or antiplatelet medications or supplements, as pau d'arco may

theoretically increase the risk of bleeding. Avoid with known allergy/hypersensitivity to pau d'arco, its constituents, or members of the Bignoniaceae family.[108]

Drug Interactions

Avoid in those who are taking anticoagulant, or antiplatelet medications or supplements, as pau d'arco may theoretically increase the risk of bleeding.

Administration and Dosage

Liquid Extract:	1:1
Alcohol:	45%
Weekly Dosage:[110]	10 to 20mL

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