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Review Article

Phytochemicals from *Phyllanthus niruri* Linn. and their pharmacological properties: a review

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Abstract

This review discusses the medicinal plant *Phyllanthus niruri* Linn. (Euphorbiaceae), its wide variety of phytochemicals and their pharmacological properties. The active phytochemicals, flavonoids, alkaloids, terpenoids, lignans, polyphenols, tannins, coumarins and saponins, have been identified from various parts of *P. niruri*. Extracts of this herb have been proven to have therapeutic effects in many clinical studies. Some of the most intriguing therapeutic properties include anti-hepatotoxic, anti-lithic, anti-hypertensive, anti-HIV and anti-hepatitis B. Therefore, studies relating to chemical characteristics and structural properties of the bioactive phytochemicals found in *P. niruri* are very useful for further research on this plant as many of the phytochemicals have shown preclinical therapeutic efficacies for a wide range of human diseases, including HIV/AIDS and hepatitis B.

Introduction

Phyllanthus is a large genus of shrubs, trees and rare herbs of the family Euphorbiaceae, comprising more than 600 species, of which *P. accuminatus*, *P. amarus*, *P. pulcher*, *P. niruroides*, *P. anisolobus*, *P. orbiculatus*, *P. emblica*, *P. oxyphyllus*, *P. flexuosus*, *P. raticulatus*, *P. fraternes*, *P. simplex*, *P. mullernus*, *P. urinaria*, *P. mytrifolis*, *P. virgatus*, *P. niruri* and *P. watsonii* were investigated for their phytochemical and pharmacological properties. The genus is found in almost over all warmer parts of the world (Burkill 1996).

Among the *Phyllanthus* species, *P. niruri* is a small erect annual herb growing up to 30–40 cm in height and is indigenous to the Amazon rainforest and other tropical areas, including South East Asia, Southern India and China (Girach et al 1994). Its leaves are 7–12 cm long and they are alternate, sessile oblong. It has small off-white-greenish flowers, which are solitary, auxiliary, pedicellate, apetalous and monoecious. *P. amarus* and *P. sellowianus* are closely related to *P. niruri* in appearance, phytochemical contents and history, but they are found in drier regions of India and Brazil, and even in Florida and Texas. In a recent report, cladistic analysis indicated that the *Phyllanthus* genus is paraphyletic and therefore the two problematic and confusing species, *P. niruri* and *P. amarus*, are two individual species (Lee et al 2006).

P. niruri as a herbal medicine

P. niruri has a long history in herbal medicine systems such as Indian Ayurveda, Traditional Chinese Medicine and Indonesian Jamu. The whole plant is used as remedies for many conditions such as dysentery, influenza, vaginitis, tumours, diabetes, diuretics, jaundice, kidney stones and dyspepsia. The plant is also useful for treating hepatotoxicity, hepatitis B, hyperglycaemia and viral and bacterial diseases (Chopra et al 1986). *P. niruri* has been used in Ayurvedic medicine for over 2000 years and has a wide number of traditional uses for jaundice, gonorrhoea, frequent menstruation and diabetes. It is an important medicinal plant in jamu, a well-known Indonesian traditional herbal medicine to treat various diseases. In jamu preparations, the plant is used as antiviral and hepatoprotective agent. In Malaysia, *P. niruri*, known as Dukong anak, is used internally for diarrhoea, kidney disorders, gonorrhoea and coughs (Burkill 1996).

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P. niruri is called Chanca Piedra in Spanish, which means stone breaker, because it has been used as an effective remedy to eliminate gallstones, kidney stones and other kidney disorders. In a preclinical study, *P. niruri* aqueous extract exhibited a potent and effective non-concentration-dependent inhibitory effect on calcium oxalate (CaOx) crystal formation (Campos & Schor 1999). This response was present even at higher concentrations. This may explain why it has long been used in traditional medicine as a preventive to kidney stone formation (Freitas et al 2002). In Brazilian herbal medicine, it is called Quebra Pedra and is considered an excellent remedy for hydropsy, urinary and bladder infections. It is also used to cure kidney disorders, hepatitis and diabetes (Santos et al 1995; Wang et al 1995; Wang 2000). In India, where it is called Pitirishi or Budhatri, it is a common household remedy for asthma, bronchitis, coughs, extreme thirst, anaemia, jaundice and tuberculosis (Dhar et al 1968).

P. niruri has been the subject of much research to investigate the active constituents and their pharmacological activity, beginning in the mid-1960s. Ottow was the first to work on *P. niruri* and reported the isolation of phyllanthin in 1891 (Row et al 1964). It has a rich source of phytochemicals, many of which have been found only in *P. niruri* (Dhar et al 1968).

Many of the active constituents to which the biological activity of *P. niruri* has been attributed include lignans, tannins, coumarins, terpenes, flavonoids, alkaloids, saponins and phenylpropanoids, which have been found in the leaves, stem and roots of this plant. Common lipids, sterols and flavonols also occur in the plant (Dhar et al 1968).

Indian and Brazilian research groups were the first to conduct studies relating to the medicinal properties of *P. niruri* since this plant is indigenous to their areas with a long history of use by their inhabitants (Unander et al 1991). Brazilian researchers documented the antispasmodic activity of an alkaloid of *P. niruri* (Calixto et al 1984). *P. niruri* gained worldwide attention in the late 1980s due to its activity against hepatitis B (Venkateswaran et al 1987).

An alkaloid extract of *P. niruri* demonstrated smooth muscle relaxation effect specific to the urinary and biliary tracts (Kitisin et al 1952). The anti-hepatotoxic activity of *P. niruri* has been attributed to two novel lignans, phyllanthin and hypophyllanthin (Symasundar et al 1985). Glycosides (quercitrin and geraniin) found in *P. niruri* demonstrated aldose reductase inhibitory (ARI) activity in studies conducted by a Japanese research group in 1988 and 1989 (Ueno et al 1988; Shimizu et al 1989). The ARI effect was also due to the presence of another ellagitannin phytochemical, ellagic acid (Shimizu et al 1989). The plant also possessed potent analgesic activity against pain models in rats (Santos et al 1994; Martini et al 2000).

The diuretic, hypotensive and hypoglycaemic effects of *P. niruri* were documented in a human study, which showed a significant diuretic effect (Devi et al 1986). Similar studies in man revealed that *P. niruri* caused reduction in the systolic blood pressure in non-diabetic hypertensive patients and reduction of blood glucose in diabetic patients (Ramakrishnan et al 1982; Hukeri et al 1988).

In-vitro and in-vivo studies showed that extracts of *P. niruri* effectively protected against liver damage induced by various chemical liver toxins (Sreenivasarao et al 1985; Thabrew et al 1996). Indian researchers reported that *P. niruri* was an effective single drug in the treatment of jaundice in children (Dixit et al 1982). In recent studies, the protein fraction of *P. niruri* demonstrated protection of liver tissues against oxidative stress in mice (Bhattacharjee & Sil 2006; Chatterjee et al 2006).

Research on *P. niruri* revealed that its antiviral activity extends to human immunodeficiency virus (HIV) and a simple aqueous extract of the plant inhibited HIV-1 reverse transcriptase (Ogata et al 1992; Naik & Juvekar 2003). A Brazilian research group reported that intake of *P. niruri* reduces urinary calcium, based on the analysis of a subset of patients presenting with hypercalciuria (Nishiura et al 2004). An inhibitory effect of *P. niruri* extract on CaOx crystal growth and aggregation in human urine suggested that it might interfere with the early stages of stone formation and represent an alternative form of prevention for urolithiasis (Barros et al 2003).

The in-vitro and in-vivo anti-plasmodial activities of the ethanolic and dichloromethane extracts, as well as the toxicity of the lyophilized aqueous extract, from *P. niruri* were also reported (Tona et al 1999; Cimanga et al 2004). The dichloromethane extract of *P. niruri* whole plant showed 100 and 81.7% inhibition of *Plasmodium falciparum* growth at concentrations of 600 and 6 $\mu g\,mL^{-1}$, respectively. At the same concentrations, the ethanol extract gave 100 and 64.5% inhibition, respectively (Tona et al 1999). The antimalarial activity of *P. niruri* was also observed in a 4-day suppressive assay against *P. berghei* ANKA in mice (Tona et al 2001). The lipid-lowering activity of *P. niruri* was also studied in triton- and cholesterol-fed hyperlipaemic rats (Khanna et al 2002).

Structures and pharmacological activities of phytochemicals

Flavonoids

Rutin Rutin (Figure 1), a flavonol glycoside comprised of the flavonol quercetin and the disaccharide rutinose, belongs to a large group of phenolic secondary metabolites of plants that includes more than 2000 different known chemicals (Chauhan et al 1977). Rutin is important because it strengthens capillaries and so is able to help people suffering from arteriosclerosis or high blood pressure (Becker et al 1985). Free radicals are said to be responsible for as much as 90% of all human diseases, such as cancer, arteriosclerosis, stroke and senility, due to ageing, and recent studies have shown that the rutins are powerful anti-oxidants that fight free radicals (Gao et al 2003).

Quercetin Quercetin (Figure 1) is important in nutrition due to its ability to strengthen and modulate the permeability of the walls of the blood vessels including capillaries (Gupta et al 1984). This bioactive compound has anti-aggregant, anticancer, anti-fungal (especially anti-dermatophytic), anti-feedant, anti-glaucomic, anti-inflammatory, anti-oxidant, antiseptic and anti-spasmodic activity (Saija et al 2003).

Quercitrin Quercitrin (quercetin 3-*O*-alpha-L-rhamnopyranoside) (Figure 1) is a flavone glycoside and has anti-leishmanial,

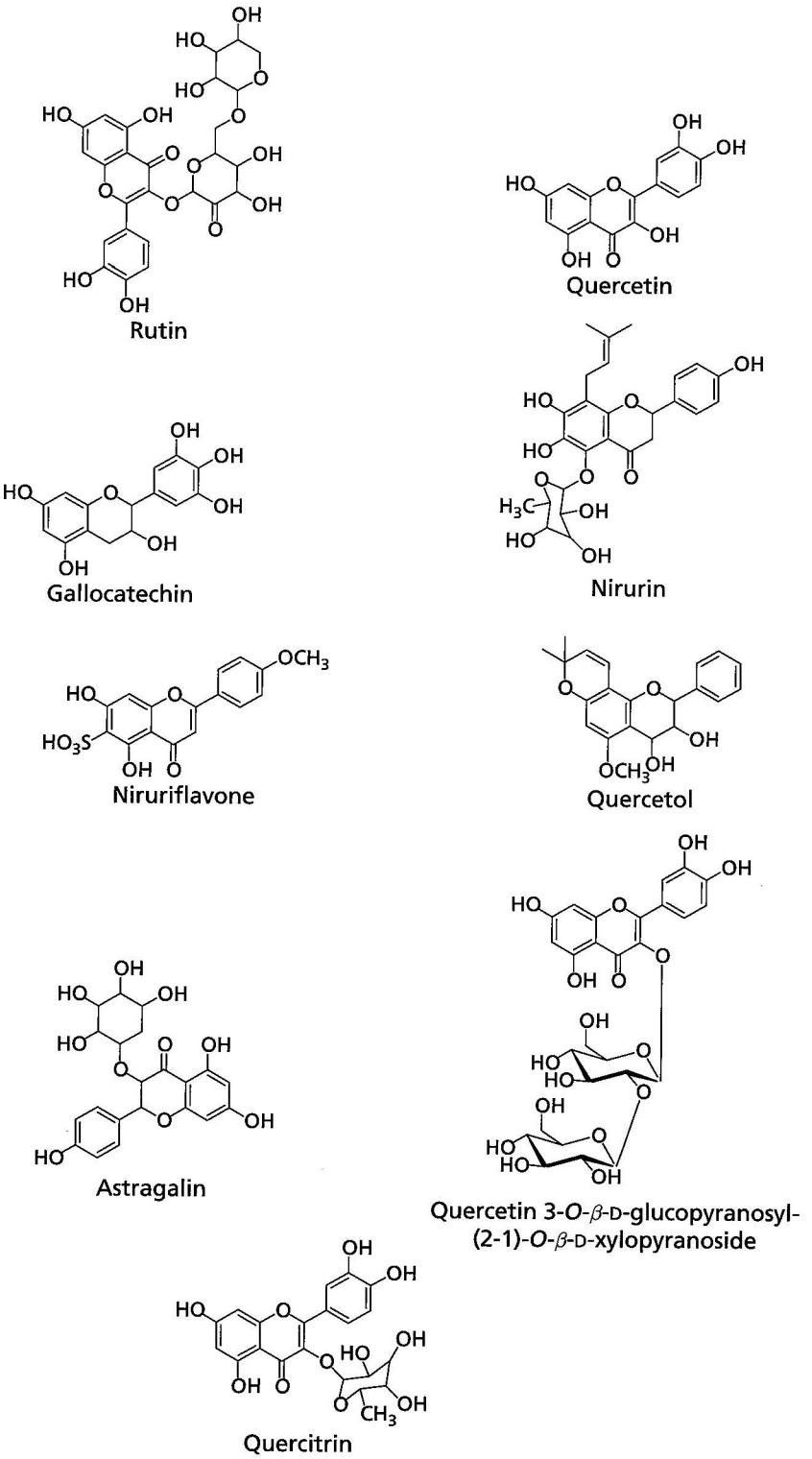


Figure 1 Chemical structures of flavonoids.

anti-nociceptive and anti-inflammatory activity (Camuesco et al 2004; Gadotti et al 2005; Muzitano et al 2006). It has demonstrated a potent anti-leishmanial activity (IC_{50} approximately 1 $\mu g\,mL^{-1}$) with a low toxicity profile (Muzitano et al 2006). Quercitrin also produced a dose-dependent inhibition of acetic-acid-induced visceral pain in mice, with a mean ID_{50} value of 2.4 $mg\,kg^{-1}$ (Gadotti et al 2005). The compound showed anti-diarrhoeal activity against castor-oil- and PGE_2-induced

diarrhoea in mice (Galvez et al 1993). It was also reported that quercitrin exerted a beneficial effect in experimental colonic inflammation (Sanchez de Medina et al 1996, 2002; Galvez et al 1997; Cruz et al 1998).

Astragalin Astragalin (Figure 1) is a flavanone and has diuretic activity (Kale et al 2001). It also has the ability to regulate the immunological capacity of man and animals by enhancing phagocytosis, increasing the number of macrophages and promoting synthesis of antibodies (Gan 1998). Astragalin inhibits passive cutaneous anaphylaxis (PCA) reactions in mice and histamine release in cultured KU812 cells (a human basophil cell line) (Kotani et al 2000). The compound has the potential to reduce the incidence and severity of dermatitis and serum concentrations of IgE in NC/Nga atopic mice (Kotani et al 2000). Reduced inflammatory cell infiltrate was noted on histological analysis of the skin of D-*N*-galactosamine-lipopolysaccharide-treated mice (Matsuda et al 2002).

Catechin Gallocatechin (Figure 1) was isolated from the tissue cultures of *P. niruri* (Ishimaru et al 1992). It was reported that catechins suppressed the growth of human colon and hepatic epithelial cancer cells and induced apoptosis (Uesato et al 2001). Catechins in the presence of trace levels of elements, in particular Cu^{2+}, have pro-oxidative activity and bactericidal effects (Nobuo et al 1999; Desong et al 2004).

Others Prenylated flavanone glycoside, nirurin (5,6,7,4′-tetrahydroxy-8-(3-methylbut-2-enyl) flavanone-5-*O*-rutinoside), and quercetol (Figure 1) were also isolated from *P. niruri* (Gupta et al 1984). Niruriflavone (Figure 1) is a flavone sulfonic acid and was shown to exhibit potent radical scavenging properties in 2, 2′-azinobis(3-ethylbenzthiazoline-6-sulfonate) (ABTS) cation radical reduction assay (Than et al 2006).

Terpenes

Limonene Limonene (Figure 2) is a monoterpenoid and has many medical and pharmaceutical applications (Singh et al 1989a). The limonene molecule exists in two forms that are mirror images of each other and it can be synthesized from two molecules of isoprene by the Diels-Alder reaction (Kurt 1983). Limonene is a major component of orange oil. The anti-carcinogenic actions of limonene in liver tumour models have been reported (Mills et al 1995). An increase in diclofenac permeation induced by limonene has also been described in hairless rats, which suggests that limonene is an effective topical medication for both dermal and sub-dermal injuries (Obata et al 1993).

p-Cymene *p*-Cymene (Figure 2) also belongs to the monoterpene category with a wide variety of biological actions, such as anti-oxidant and anti-microbial activity (Allardyce et al 2003). In a recent study by Tomaino et al (2005), thyme, an essential oil with *p*-cymene as one of its components, exhibited a good anti-oxidant activity by preventing oxidation of α-tocopherol. *p*-Cymene was tested for anti-microbial properties using the paper disc diffusion method, in which it revealed a good anti-microbial activity (Medeiros et al 2003). Synthetic derivatives of *p*-cymene, such as chloro (*p*-cymene) ruthenium complex compounds, displayed in-vitro anti-cancer activity against human mammary cancer cell line (MDA-MB-435) (Huxham et al 2003).

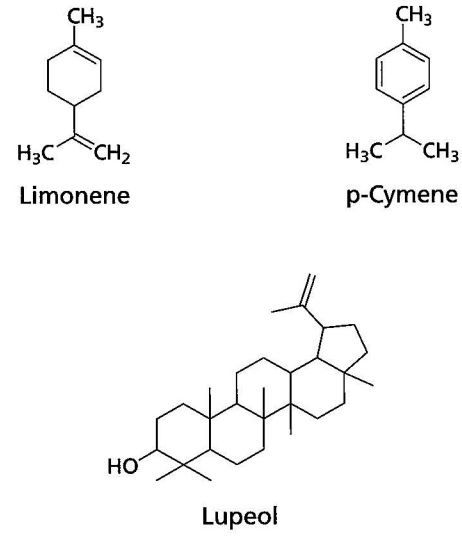


Figure 2 Chemical structures of terpenoids.

Lupeol Lupeol (Figure 2) belongs to the pentacyclic triterpene category and can be synthesized through protosterol carbocation by backbone rearrangement reaction (Chauhan et al 1976). It suppresses superoxide generation by preventing tyrosyl phosphorylation of a 45 kDa protein in human neutrophils (Yamashita et al 2002). The compound has proven to have anti-inflammatory and anti-tumour activities (Geetha & Varalakshmi 2001; Saleem et al 2005). More interestingly, lupeol and its derivatives have been shown to inhibit CaOx crystal aggregation in experimental urolithiasis (Malini et al 2000), mimicking the activity of the whole-plant extract (Campos & Schor 1999). Lupeol, which is an effective skin chemopreventive agent, can suppress benzoyl-peroxide-induced cutaneous toxicity (Saleem et al 2001). It also has the ability to improve the antioxidant status of liver against cadmium-induced toxicity in rats (Sunitha et al 2001).

Coumarins

Ellagic acid Ellagic acid (Figure 3) is a phenolic compound that has potent anti-carcinogenic and anti-viral properties (Rice-Evans et al 1996). Ellagic acid itself is not thought to be naturally present in plants. Instead, polymers of gallic acid and hexahydroxydiphenoyl (HHDP) are linked to glucose centres to form a class of compounds known as ellagitannins (Rice-Evans et al 1996). When two gallic acid groups become linked side-by-side within a tannin molecule, the HHDP group is formed. Ellagic acid is the result when the HHDP group is cleaved from the tannin molecule and spontaneously rearranges. The availability of ellagic acid to the body from dietary sources has only been confirmed with red raspberries (Ueno et al 1988; Ferguson 2001). Ellagic acid

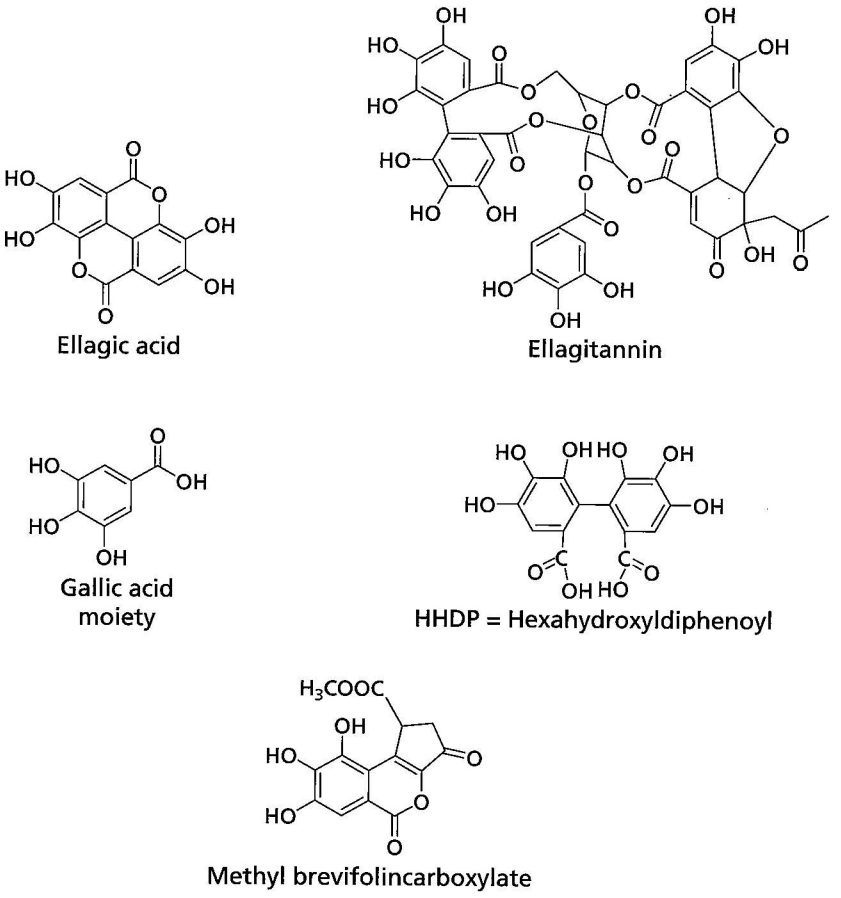


Figure 3 Chemical structures of ellagic acid, gallic acid, elligitannin, hexahydroxyldiphenoyl moiety and methyl brevifolincarboxylate.

from red raspberries was found to prevent the binding of carcinogens to DNA and reduce the incidence of cancer in cultured human cells exposed to carcinogens (Whitley et al 2003). The cancer chemopreventive effect of ellagic acid was related to its ability to scavenge cancer-causing chemicals, making them inactive. It also inhibited the ability of other chemicals to cause mutations in bacteria (Whitley et al 2003). Some articles in which ellagitannins are quantified refer to ellagic acid because quantification of ellagitannins is done by breaking them down into ellagic acid subunits and then quantifying the subunits (Whitley et al 2003).

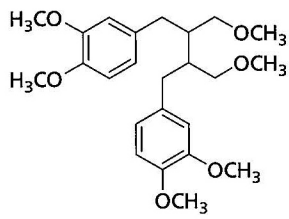
Methyl brevifolincarboxylate Methyl brevifolincarboxylate (Figure 3), a coumarin derivative isolated from the leaves of *P. niruri*, showed a vasorelaxant effect on rat aortic rings. This compound inhibited noradrenaline-induced vasocontractions, which was in part attributable to a decrease in Ca^{2+} concentration, through receptor-operated Ca^{2+}-channels (Iizuka et al 2006).

Lignans

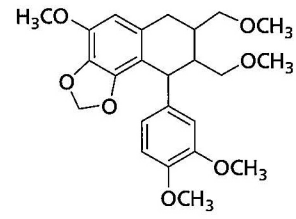
Phyllanthin and hypophyllanthin (Figure 4) belong to the lignan category and have been shown to possess hepatoprotective and anti-genotoxic activities (Row et al 1964). The two compounds exhibited a significant protection against CCl_4- and galactosamine-induced elevation in liver transferase enzymes levels and significantly increased protein levels (Symasundar et al 1985). These compounds were also reported to inactivate hepatitis B, both in-vitro and in-vivo (Venkateswaran et al 1987). Phyllanthin, a lignan of the aryltetrahydronaphthalene type, possesses anti-viral activity against HIV by inhibiting reverse transcriptase (Sagar et al 2004).

Niranthin, nirtetralin, phyltetralin and lintetralin (Figure 4) were also isolated from *P. niruri* (Anjaneyulu et al 1973; Ganeshpure et al 1981; Satyanarayana & Venkateswarlu 1991). Phyltetralin, nirtetralin and niranthin exhibited anti-inflammatory activity by inhibiting carrageenan-induced paw oedema and neutrophil influx (Kassuya et al 2005). Huang et al (2003) reported the anti-HBsAg and anti-HBeAg activities of niranthin and nirtetralin and the compounds suppressed HBsAg and HBeAg expression effectively at a non-cytotoxic concentration of 50 μM.

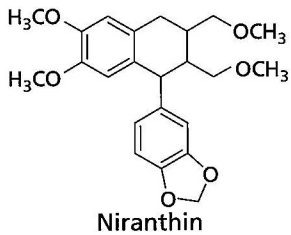
Isolintetralin, 2,3-desmethoxy seco-isolintetralin, 2,3-desmethoxy seco-isolintetralin diacetate, linnanthin, demethylenedioxyniranthin, nirphyllin and phyllnirurin (Figure 4) were also reported from *P. niruri* (Singh et al 1989b; Satyanarayana & Venkateswarlu 1991). Satyanarayana et al (1988) reported the isolation of an unusual seco lignan, seco-4-hydroxylintetralin, two hydroxy-lignans, seco-isolariciresinol trimethyl ether and hydroxyniranthin, and a known dibenzylbutyrolactone 3,4-



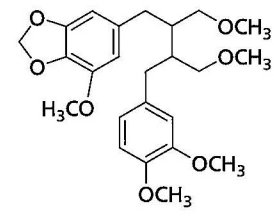
Phyllanthin



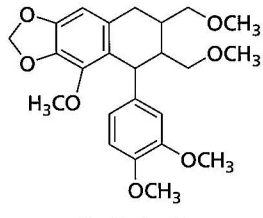
Hypophyllanthin



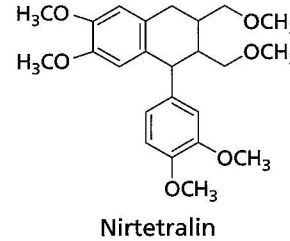
Niranthin



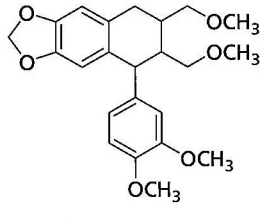
Lintetralin



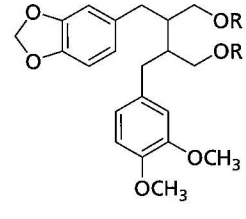
Phyltetralin



Nirtetralin



Isolintetralin



R = H 2,3-Desmethoxy seco-isolintetralin
R = $COCH_3$ 2,3-Desmethoxy seco-isolintetralin diacetate

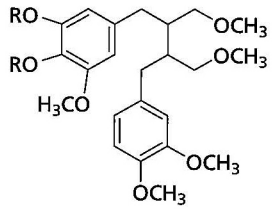
Figure 4 Chemical structures of lignans.

methylenedioxybenzyl-3′,4′-dimethoxybenzylbutyrolactone from the leaves of *P. niruri*. 3,4-Methylenedioxybenzyl-3′,4′-dimethoxybenzylbutyrolactone was previously isolated from *Bursera schlechtendalii* and reported to exhibit anti-tumour activity (McDoniel & Cole 1972). There are no pharmacological studies reported on most of the above-mentioned phytochemicals.

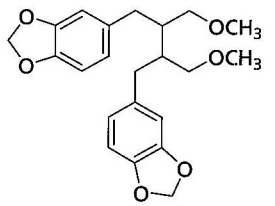
Recently, Elfahmi et al (2006) reported the isolation of two new lignans, namely cubebin dimethyl ether and urina tetralin from the cell suspension cultures of *P. niruri*, and this was the first report on cell suspension cultures of *P. niruri* that successfully produced lignans. Urinatetralin was previously isolated from *P. urinaria* (Chang et al 2003).

Tannins

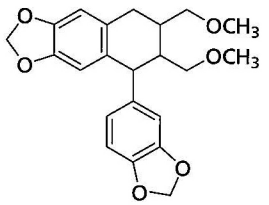
Repandusinic acid Repandusinic acid (Figure 5) inhibited human immunodeficiency virus type-1 reverse transcriptase (HIV-1-RT) with an ID_{50} value of 0.05 μM (Ogata et al 1992). It showed 10-fold more sensitivity towards HIV-1-RT compared with human DNA polymerase-α, which is an indication of good selectivity in inhibiting HIV replication without harming normal cells of the body. In addition, 4.5 μM of this compound inhibited 50% of HIV-1-induced giant cell formation (Ogata et al 1992). In the same study, this compound at 2.5 μM inhibited HIV-1 specific p24 antigen production in Clone H9 cell system. The compound also showed strong inhibitory activity against HIV-1 protease, with an IC_{50} value of 12.5 μM (Xu et al 2000).



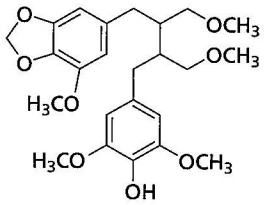
R = CH_3 linnanthin
R = H Demethylenedioxyniranthin



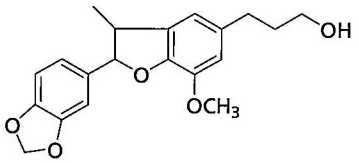
Cubebin dimethyl ether



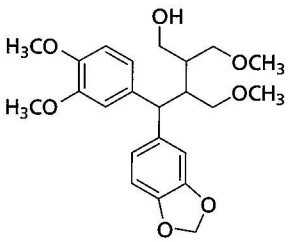
Urinatetralin



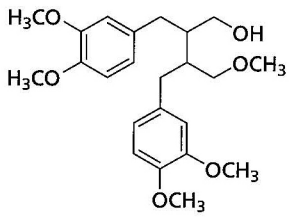
Nirphyllin



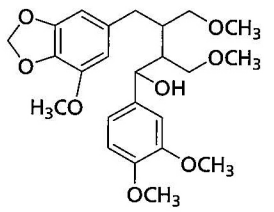
Phyllnirurin



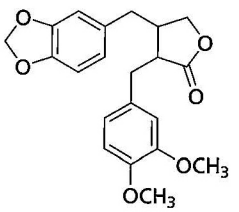
Seco-4-hydroxylintetralin



Seco-isolariciresinol trimethyl ether



Hydroxyniranthin



3,4-Methylenedioxybenzyl-3',4'-dimethoxybenzylbutyrolactone

Figure 4 (Continued).

Geraniin Geraniin (Figure 5) isolated from *P. niruri* (Ueno et al 1988) exhibited significant and dose-related anti-nociceptive property against acetic-acid-induced abdominal constrictions in mice. It was shown that geraniin was 6- to 7-fold more potent at the ID_{50} level (μmol kg^{-1}) compared with aspirin and paracetamol (Miguel et al 1996). It also possessed the ability to lower systemic blood pressure through the reduction of noradrenaline release in spontaneously hypertensive rats (Cheng et al 1994).

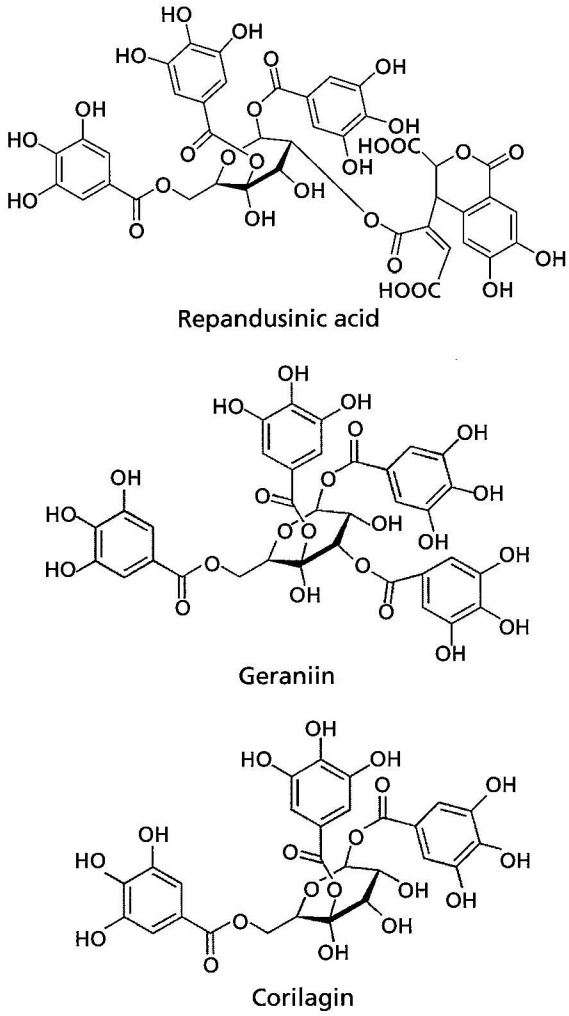


Figure 5 Chemical structures of tannins.

Corilagin Corilagin also found in *P. niruri* (Shimizu et al 1989). Corilagin significantly inhibited plasminogen-activator-inhibitor-1 (PAI-1) activity in rat plasma or platelet-released substances while it elevated plasma tissue-type plasminogen (tPA) activity, in a concentration-dependent manner, and this property is assumed to be responsible for its thrombolytic effect (Shen et al 2003). The compound possesses the ability to lower blood pressure through the reduction of noradrenaline release and direct vasorelaxation in the spontaneously hypertensive rat (Cheng et al 1995). Corilagin was also evaluated for its anti-fungal activity against *Candida glabrata* strains, in which it showed a minimum inhibitory concentration (MIC) of 0.8 nM, similar to amphotericin B (MIC 0.5 nM) and sertaconazole (MIC 0.9 nM) (Latte & Kolodziej 2000).

Saponins

Saponins are high-molecular-weight glycosides combining a sugar and a steroid aglycone or triterpene molecule. Diosgenin (Figure 6) is a steroid glycoside (Kurt 1983) that exists widely in the plant kingdom and plants containing this agent are intensively used in Traditional Chinese Medicine. Diosgenin demonstrated anti-fungal and cardiovascular activity (Hufford et al 1988; Au et al 2004).

Alkaloids

Norsecurinine Norsecurinine (Figure 7) is a securinega type alkaloid and has strong anti-spasmodic activity (Joshi et al 1986). Structurally related securinine exhibits good anti-malarial and anti-bacterial activities (Mensah et al 1990; Weenen et al 1990). Equilibrium binding assays revealed that securinine inhibits GABA (gamma-aminobutyric acid) binding to its receptor with an IC_{50} of approximately 50 μM (Beutler et al 1985). Extracellular electrophysiological studies on neurons in the cat spinal cord indicated that securinine blocked the inhibitory action of GABA while having no effect on that of glycine. These results suggested that securinine is a selective antagonist of GABA recognition sites on mammalian central neurons (Beutler et al 1985). Nirurine, phyllanthine and phyllochrysine (Figure 7) (Cuellar & Estevez 1980; Bunyapraphatsara et al 1983; Mulchandani et al 1984) were also isolated from *P. niruri*.

Other compounds

A recent study reported the presence of 1-*O*-galloyl-6-*O*-luteoyl-α-D-glucose and β-glucogallin in *P. niruri* (Figure 8) (Subeki et al 2005). These compounds showed good anti-babesial

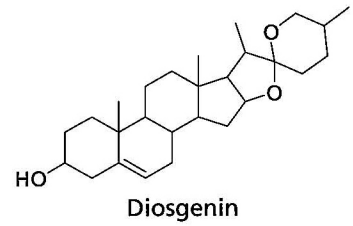


Figure 6 Chemical structure of diosgenin.

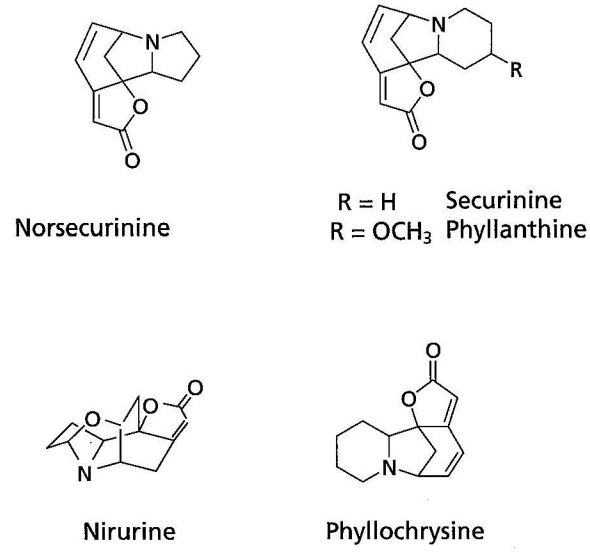
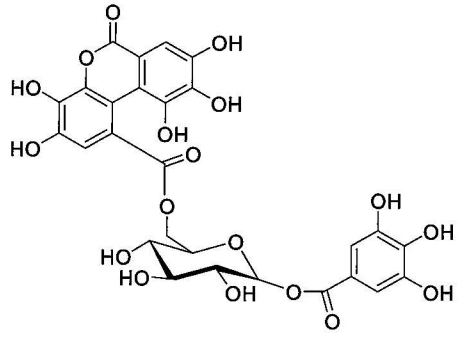
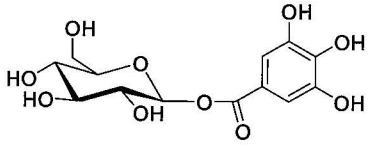


Figure 7 Chemical structures of alkaloids.

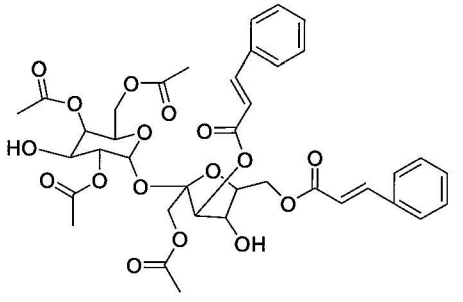
activity by inhibiting the intra-erythrocytic parasite, *Babesia gibsonii* with IC_{50} values of 4.7 $\mu g\,mL^{-1}$ and 7.5 $\mu g\,mL^{-1}$, respectively. This parasite causes canine babesiosis (haemolytic disease). These compounds were also active against the malaria-causing *Plasmodium falciparum* with IC_{50} values of 1.4 $\mu g\,mL^{-1}$ and 4.6 $\mu g\,mL^{-1}$, respectively (Subeki et al 2005). Linear and complex acidic hetero xylans were also isolated from *P. niruri* (Mellinger et al 2005a). These xylans have immunomodulatory and anti-tussive properties (Ebringerova et al 2002; Kardosova et al 2002). An aqueous extract of *P. niruri* contained an acidic arabinogalactan, which belongs to the polysaccharide category, and has shown immunostimulatory activity by stimulating macrophages from mouse peritoneal cavity to produce superoxide (O_2^-) (Mellinger et al 2005b). This type of agent, collectively known as biological response modifiers, are able to mediate many therapeutic effects, including anti-tumour activity. Niruriside (Figure 8), a phenyl propanoid, displayed anti-HIV activity by specifically inhibiting the binding of REV (regulation of virion expression) viral protein to RRE (REV-responsive element) RNA, for REV functioning. REV regulates the transport of viral RNA to the cytoplasm and, thus, is essential for HIV productive infection. The compound had an IC_{50} value of 3.3 μM for inhibition of REV–RRE complex formation (Qian-Cutrone et al 1996). Triacontanal and tricontanol (long-chain hydrocarbons) were also isolated from *P. niruri*. Triacontanal was shown to be protective against galactosamine-induced toxicity in rat hepatocytes primary culture (Symasundar et al 1985).



1-*O*-galloyl-6-*O*-luteoyl-α-D-glucose



β-Glucogallin



Niruriside

Figure 8 Chemical structures of 1-*O*-galloyl-6-*O*-luteoyl-α-D-glucose, β-glucogallin and niruriside.

Conclusions

P. niruri has many effective traditional uses for a wide variety of diseases. Some of the medicinal usages have been proven in experimental models, which suggest that the extracts of the plant possess various pharmacological actions. Owing to the impressive preclinical therapeutic potential, the plant extracts have been evaluated in human trials for the treatment of hypertension, jaundice, diabetes, hypercalciuria and urolithiasis. Subsequent studies revealed the preclinical pharmacological activity and therapeutic potential of phytochemicals isolated from *P. niruri*. The phytochemicals exhibited different structural characteristics with various pharmacological actions: lignans had excellent hepatoprotective and anti-viral properties, whereas terpenes exhibited anti-cancer, as well as anti-microbial activity. Flavonoids from *P. niruri* showed anti-oxidant activity and the alkaloids exhibited anti-spasmodic activity. More importantly, there have been no side effects or toxicity reports from many years of research on this herb. Although there has been extensive research on this herb, there is still a lot of scope for further research, especially towards the mechanism of biological activity of phytochemicals from *P. niruri* with emphasis on agents with anti-HIV and anti-hepatitis B properties. Mechanisms studies are expected to lead the way in the discovery of new agents with improved and intriguing pharmacological properties. This could be achieved by molecular modelling studies involving interaction of bioactive phytochemicals from *P. niruri* with their respective molecular targets. Upon improvement of binding affinity to the specified target by virtual chemical modification of existing pharmacophores, new small molecules could then be identified and synthesized in the laboratory. This type of study has yielded HIV protease inhibitors as therapeutic agents for the treatment HIV/AIDS (Roberts et al 1990; Kalish et al 1995).

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