PHYTOTHERAPY RESEARCH
Phytother. Res. **29**: 818–824 (2015)
Published online 20 March 2015 in Wiley Online Library
(wileyonlinelibrary.com) **DOI**: 10.1002/ptr.5335

REVIEW

Antioxidant, Anti-inflammatory, and Chemoprotective Properties of *Acacia catechu* Heartwood Extracts

Sidney J. Stohs[1,2]* and Debasis Bagchi[2,3]

[1]School of Pharmacy and Health Professions, Creighton University Medical Center, Omaha 68178, NE, USA
[2]AdvoCare International, Plano 75074, TX, USA
[3]Department of Pharmacological and Pharmaceutical Sciences, University of Houston College of Pharmacy, Houston, TX, USA

Aqueous extracts of *Acacia catechu* heartwood are rich source of catechin and epicatechin (gallic acid derivatives), with smaller amounts of flavonoids. Extracts have also been prepared with ethyl acetate, ethanol, and methanol, and the properties of these extracts have been studied and are reviewed. Potent antioxidant activity has been well established in both *in vitro* and *in vivo* studies. This antioxidant activity is believed to be responsible for the anti-inflammatory, tissue protectant, antineoplastic, and analgesic activities that have been demonstrated and clearly established in animal and cell culture systems. Furthermore, antihyperglycemic, antidiarrheal, antinociceptive, and antipyretic activities have been demonstrated in animal studies. No adverse effects have been observed in animal or human studies or in cell culture systems. In spite of the fact that *Acacia* products have been used for many years and the general safety of catechins and epicatechins is well documented, few human studies have ever been conducted on the efficacy or safety of *A. catechu* heartwood extracts. Several studies have shown that a two-ingredient combination product containing *A. catechu* extract exhibited no adverse effects when administered daily for up to 12 weeks while exhibiting significant anti-inflammatory activity in subjects with osteoarthritis of the knee. There is a need for additional human clinical studies with regard to efficacy and safety. © 2015 The Authors. Phytotherapy Research published by John Wiley & Sons Ltd.

Keywords: Acacia catechu; heartwood extracts; antioxidant; anti-inflammatory; chemoprotective; antinociceptive; catechins; epicatechins; flavonoids.

INTRODUCTION

There are over 1300 species of *Acacia* (family Mimosaceae), and exudates, leaves, seeds, heartwood, and bark are used in numerous ways. Gum Acacia (gum arabic) has been used for its medicinal properties for approximately 2500 years. It is employed as a demulcent (soothing and protectant agent) and emulsifier, and is used to increase viscosity of solutions and suspensions. It is also used in the food, soft drink, textile, tanning, cosmetic, and confection industries. As a consequence, wide exposure to gum Acacia occurs.

In recent years, numerous studies have examined various pharmacological properties of extracts prepared from heartwood, bark, leaves, seeds, and seed pods of *Acacia* species. This review will focus on the published studies associated with *Acacia catechu* Willd (Mimosaceae) heartwood extracts, which historically have been known for their high tannin content that is obtained by boiling the heartwood with water and evaporating the solution to prepare the extract. *Acacia catechu* extracts have also played a role in chemistry, with various names of chemicals as catechin, catechol, and catecholamine being derived therefrom.

* Correspondence to: Sidney J. Stohs, School of Pharmacy and Health Professions. Creighton University Medical Center, Omaha 68178, NE, USA.
E-mail: sid.stohs9@gmail.com

Acacia catechu is also known as kattha (Urdu), khadir (Hindustani and Punjabi), khoyer(Bengali and Assamese), khair and babul (Hindi), kaath (Marathi), and kachu (Malay). It is indigenous in India, other Asian countries, and East Africa. Traditionally, *A. catechu* has been used as an antimicrobial, anti-inflammatory and antifungal, coagulant, vermifuge, antidiarrheal, and astringent, and has also been employed to heal wounds, treat obesity and diabetes, and maintain oral hygiene, (Chauhan *et al.*, 2011).

Acacia catechu heartwood extracts have also been used traditionally in the preparation of betel quid (paan masala), which consists of *Piper betle* leaves, *A. catechu* paste, chopped *Areca* nut, lime, and various spices with or without tobacco (Thomas and Kearsley, 1993; Rahmatullah *et al.*, 2013; Waris and Nagi, 2014). Betel chewing is used to produce euphoria, a sense of well-being, heightened sense of alertness, and psychostimulation (Chu, 2001). This review primarily focuses on applications of *A. catechu* heartwood extracts other than its use in conjunction with betel quid.

CHEMISTRY

High-performance liquid chromatography coupled with electrospray ionization mass spectroscopy of an aqueous extract of the heartwood and leaves of *A. catechu* has

Received 16 November 2014
Revised 20 February 2015
Accepted 25 February 2015

shown that the primary constituents are catechins (Shen *et al.*, 2006), which by definition are gallic acid (polyhydroxylated benzoic acid) derivatives and polymers. The predominant catechins in *A. catechu* include catechin, epicatechin, epicatechin-3-*O*-gallate, and epigallocatechin-3-*O*-gallate. Other major secondary products present in the extracts included flavonol glycosides, flavonal dimers, and caffeine.

Other constituents that have been identified in aqueous extracts of *A. catechu* include rhamnetin, 4-hydroxyphenol, 3,3′,5,5′,7-pentahydroxyflavane, fisetinidol, 5-hydroxy-2-[2-(4-hydroxyphenyl)acetyl]-3-methoxybenzoic acid, and (2S,3S)-3,7,8,3′,4′-pentahydroxyflavane (Li *et al.*, 2011). The presence of high amounts of gallic acid-derived compounds is primarily responsible for the astringent, tanning, and antioxidant properties of the extracts.

An extract from the heartwood of *A. catechu* was shown to contain 66.9% catechin and 23.1% epicatechin; thus, 90% of the composition of this extract consisted of these two components (Hiraganahalli *et al.*, 2012). Sulaiman and Balachandran (2012) reported that an *A. catechu* powdered sample is high in gallic acid-related compounds, containing approximately 78 mg gallic acid equivalents per gram as determined by the Folin–Ciocalteu assay.

Hazra *et al.* (2013) conducted a phytochemical screening of a 70% methanol extract of *A. catechu* heartwood. The extract was analyzed for the presence of carbohydrates, alkaloids, tannins, ascorbate, terpenoids, triterpenoids, anthraquinones, saponins, and glycosides. The extract contained measurable amounts of tannins (catechins) (20 mg catechin equivalent/100-mg extract), alkaloids, ascorbic acid, and carbohydrates, and also tested positive for terpenoids and triterpenoids as well as glycosides. No anthraquinones or saponins were detected.

HUMAN STUDIES

Although *A. catechu* extracts have been widely used for hundreds of years, few published studies involving humans exist. One of the earliest known clinical studies involved an evaluation of *A. catechu* in the treatment of lepromatous leprosy (Ojha *et al.*, 1969).

Several human studies have assessed the anti-inflammatory effects of a combination of *A. catechu* extract of heartwood, roots, or bark in combination with a root extract of *Scutellaria baicalensis* (Chinese skullcap), which has been reviewed by Bitto *et al.* (2014). The primary studies will be summarized later in the text. A 12-week placebo-controlled, double-blind, randomized safety study was conducted on 59 subjects with moderate osteoarthritis of one knee (Morgan *et al.*, 2009). The product (flavocoxid) that was used consisted of a proprietary blend of the flavonoids and flavanes of the bark of *A. catechu* and the root of *S. baicalensis*, and was given at a dose of 250 mg twice a day versus the placebo. At the end of the study, there were no significant differences with respect to laboratory values including blood chemistries and serology, blood pressure, or body systems, indicating a high degree of safety of the product at this dose. No efficacy data or information was reported.

In a subsequent study (Levy *et al.*, 2010), the *A. catechu* plus *S. baicalensis* extract product (flavocoxid) was assessed for its safety and efficacy in 220 subjects with moderate-to-severe osteoarthritis of the knee using a 12-week double-blind, randomized protocol. The subjects received either the extract product orally or naproxen (both products 500 mg twice daily). At the conclusion of the study, the authors concluded that the botanical extract product was as effective as naproxen in managing the symptoms and signs of osteoarthritis of the knee. Furthermore, the botanical product containing *A. catechu* exhibited better gastrointestinal, renal, and respiratory safety than naproxen.

Another group of investigators (Arjmandi *et al.*, 2014) conducted a 1-week randomized, double-blind clinical trial in 79 moderately osteoarthritic subjects (ages 40–90 years) to examine the efficacy of UP446, a proprietary blend of *A. catechu* and *S. baicalensis* extracts (500 mg/day), to ameliorate knee joint pain and mobility as well as selected biomarkers of inflammation in comparison to naproxen (440 mg/day). Pain, knee range of motion, and overall physical activity were evaluated in all groups. Serum interleukins 1β (IL-1β), interleukin 6 (IL-6), tumor necrosis factor-α (TNF-α), C-reactive protein, and hyaluronic acid were assessed in the fasting blood sample. The combination product exhibited significant efficacy in perceived pain, stiffness, and knee range of motion. This short clinical investigation indicates that this combination product is efficacious in ameliorating the symptoms of knee osteoarthritis.

IN VITRO ANTIOXIDANT ACTIVITY

A number of studies have examined the antioxidant activity of *A. catechu* extracts *in vitro*. Naik *et al.* (2003) demonstrated that aqueous extracts of various plant parts were capable of inhibiting radiation induced-lipid peroxidation in a rat liver microsomal preparation. Antioxidant activity was also demonstrated against 2,2-diphenyl-1-picrylhydrazyl (DPPH) radicals and radical formation by pulse radiolysis.

The antioxidant, iron-chelating, and DNA-protective properties of a 70% methanolic extract of *A. catechu* heartwood were assessed (Hazra *et al.*, 2010). The extract was shown to exhibit free radical-scavenging activity against superoxide, nitric oxide, peroxynitrite, hydrogen peroxide, singlet oxygen, and hypochlorous acid radicals. The extract used had a gallic acid phenolic equivalent of 97 mg/mL and a quercetin equivalent flavonoid content of 383.7 mg/mL.

Guleria *et al.* (2011) conducted a detailed analysis of the antioxidant activities and ability to protect against DNA strand breaks with methanol, acetone, and ethyl acetate extracts of heartwood, bark, and leaves of *A. catechu.* The results demonstrated that methanol and ethyl acetate extracts of heartwood exhibited potent antioxidant and DNA-protective activities. A heartwood extract was shown to be effective against DPPH radicals, superoxide anions, and hydroxyl radicals.

Other studies have demonstrated *in vitro* antioxidant and free radical-scavenging activities of aqueous extracts of *A. catechu* (Patil *et al.*, 2003; Kumar *et al.*, 2013), *A. catechu* bark extracts (Sulaiman *et al.*, 2011), and ethyl acetate extracts of the whole plant based on anti-lipid peroxidative activity, superoxide anion-scavenging activity, and reducing power (Noorani *et al.*, 2010). Thus, the antioxidant and radical-scavenging

activities of heartwood, leaf, and bark extracts are well established.

Methanol and hexane extracts of *A. catechu* bark were found to be more antiproliferative and cytotoxic than aqueous extracts against various cancer cell lines *in vitro* (Nadumane, 2011). At the same time, the extracts appeared to be quite safe against human peripheral lymphocytes, suggesting that the results could be extrapolated to humans.

Ghate *et al.* (2014) assessed the *in vitro* anticancer and apoptosis efficacy of a 70% methanolic extract of *A. catechu* heartwood in the cultured MCF-7 human breast adenocarcinoma cell line. The extract exhibited significant cytotoxicity towards cultured MCF-7 cells (IC_{50} (50% inhibitory concentration) = 289 μg/mL), as well as induced apoptosis as demonstrated by flow cytometric analysis and morphological investigation. Immunoblot analysis confirmed that apoptosis induction by the extract was achieved by enhancing Bax/Bcl-2 ratio with activation of the caspase cascade and ultimate cleavage of poly adeno ribose polymerase.

ANIMAL STUDIES

A number of early studies were conducted on various *A. catechu* extracts and products. The effect of administering *A. catechu* on niacin, ascorbic acid, and riboflavin status in rats was examined (Chaudhari and Hatwalne, 1970), followed by a study on the effect of *A. catechu* on niacin biosynthesis and requirements in rats (Chaudhari and Hatwalne, 1971). The hypoglycemic activity of *A. catechu* seeds in the diet was also studied (Singh *et al.*, 1976). An aqueous extract of *A. catechu* heartwood was shown to produce a dose-dependent decrease in blood pressure in both anesthetized dogs and rats (Sham *et al.*, 1984).

The hypoglycemic, hepatoprotective, antipyretic, and antidiarrheal activities of an ethyl acetate extract of *A. catechu* heartwood were demonstrated in rats (Ray *et al.*, 2006). A dose of 250 mg/kg orally exhibited significant antidiarrheal properties, while doses of 250 and 500 mg/kg significantly reduced blood sugar levels in alloxan-induced diabetic rats. Similar doses exhibited antipyretic activity as well as significant protection against carbon tetrachloride-induced liver damage. The antihyperglycemic activity of an *A. catechu* ethanolic bark extract has also been demonstrated in alloxan-induced diabetic rats (Jarald *et al.*, 2009).

Bhatia *et al.* (2011) have also shown that an ethanol extract of *A. catechu* hardwood exhibits antihyperglycemic activity in streptozotocin-induced diabetic rats at a dose of 250 mg/kg. Furthermore, the extract showed marked inhibition of aldose reductase enzyme activity in the eye lens of normal rats. In diabetic animals, glucose is metabolized by this enzyme to sorbitol and fructose, which are believed to contribute to the microvascular complications associated with diabetes.

An ethyl acetate extract of black catechu, a product obtained from *A. catechu* heartwood, at doses of 250 and 500 mg/kg, was shown to significantly reduce glucose absorption when given orally to rats (Gunindro *et al.*, 2013). The presence of tannins and flavonoids in the product were believed to be responsible for this action. This study represents another mechanism whereby *A. catechu* extract can control blood sugar levels.

In another investigation, Rahmatullah *et al.* (2013) evaluated the dose-dependent antidiabetic and antinociceptive efficacy of an aqueous extract of the heartwood of *A. catechu* in mice at doses up to 400 mg/kg body weight. A reduction of 37% in serum glucose levels was observed in an oral glucose tolerance test at a dose of 200 mg/kg body weight of the extract. A similar result was seen with a 400 mg/kg dose. In a parallel experiment, the antihyperglycemic drug glibenclamide (10 mg/kg body weight) lowered serum glucose level by 48.6%.

In the antinociceptive efficacy study, an approximately 70% reduction in writhing, induced in mice by intraperitoneal administration of acetic acid, was observed with a dose of 400 mg/kg body weight of the extract (Rahmatullah *et al.*, 2013). In a parallel experiment, the standard antinociceptive drug aspirin at 400 mg/kg body weight reduced the number of writhing by 60.5%. This study indicates that *A. catechu* heartwood extract is efficacious in reducing blood sugar levels in diabetic mice as well as in the alleviation of pain. The components of the extract responsible for these effects were not determined.

Several studies have demonstrated that ethyl acetate extracts of *A. catechu* heartwood when given intraperitoneally can protect against carbon tetrachloride-induced liver toxicity (Jayasekhar *et al.*, 1997; Valte *et al.*, 2012). In both studies, doses of the extract were given at 250 mg/kg. In neither case was the effect of oral administration determined.

In another study (Hiraganahalli *et al.*, 2012), the hepatoprotective and antioxidant activity of a standardized extract of *A. catechu* was evaluated against *t*-butyl hydroperoxide-induced toxicity in a human hepatocarcinoma cell line. The extract was shown to have significant hepatoprotective activity. Furthermore, the extract was shown to exhibit high antioxidant radical-quenching activity when tested *in vitro* against four different assay systems. These authors also concluded that the *A. catechu* heartwood extract has a hepatoprotectant ability, and that this activity was mainly attributable to the antioxidant activity, which reduced lipid peroxidation and cellular damage.

Hazra *et al.* (2013) investigated the dose-dependent efficacy of a 70% methanol extract of *A. catechu* heartwood (50, 100, and 200 mg/kg body weight orally) against iron-induced hepatotoxicity in Swiss albino mice over a period of 21 days. Iron-induced hepatoxicity was induced using five intraperitoneal doses (one dose every 2 days) of iron–dextran solution at 100 mg/kg body weight. Three doses of heartwood extract and a standard oral iron-chelating drug Desirox (20 mg/kg body weight) were given to various groups of mice. The heartwood extract exhibited dose-dependent reductions in hepatic iron, lipid peroxidation, protein carbonyl, liver fibrosis, serum enzymes, and ferritin. The antioxidant enzyme levels including superoxide dismutase, catalase, and glutathione S-transferase as well as reduced glutathione were enhanced, and the reductive release of ferritin iron increased significantly with gradually increasing concentrations of the heartwood extract.

Furthermore, iron overload caused damage to the hepatocellular organelles including mitochondria and lysosomes, and a significant increase in hydroxyproline, a biomarker of hepatic fibrogenesis, was observed (Hazra *et al.*, 2013). The heartwood extract significantly reduced hydroxyproline levels, demonstrating its hepatic fibrosis

inhibitory potency. Moreover, the iron induced dramatic increases in alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase, and bilirubin, all of which were significantly attenuated by the heartwood extract in a dose-dependent manner. The authors also conducted a phytochemical screening of the extract as described in the Section on Chemistry.

Monga *et al.* (2011, 2013) have conducted a series of studies on the chemoprotective effects of *A. catechu* hardwood extract in mice. In each of these studies, 7,12-dimethylbenz[a]anthracene (DMBA) or DMBA plus 12-*O*-tetradecanoylphorbol-13-acetate was used to induce tumors in the mice. In the initial study, various solvents were used with a standardized aqueous extract being further tested because of the results of cytotoxicity testing on a human epithelial carcinoma cell line (Monga *et al.*, 2011). The results showed that DMBA-induced squamous cell carcinomas in mice were inhibited by the aqueous extract in a dose-dependent manner with maximum tumor incidence being decreased by 70%.

In a second study, Monga *et al.* (2013) evaluated the *A. catechu* heartwood extract against a human breast adenocarcinoma cell line and DMBA-induced mammary carcinoma in mice. The extract was shown to exhibit a dose-dependent, potent antitumor activity. Furthermore, the extract increased the activities of the antioxidant enzymes superoxide dismutase, catalase, glutathione peroxidase, glutathione transferase, and glutathione reductase. It also increased reduced glutathione content while inhibiting lipid peroxidation. The results suggested that the antineoplastic activity of the *A. catechu* extract was related to its antioxidant activity.

In a third study, Monga *et al.* (2012) assessed the chemoprotective activity of *A. catechu* heartwood extract against DMBA-induced hepatocarcinogenesis in mice. The extract reduced the liver tumor incidence by 63.5%, and modulated the various antioxidant defense systems described earlier. The authors concluded that the catechin-rich extract exerts a chemoprotective effect by promoting the antioxidant defense system and inhibiting lipid peroxidation, and that these processes are linked to the modulation of transcription factor expression during hepatocarcinogenesis.

The conclusion from the three studies of Monga *et al.* (2011, 2012, 2013) is that an aqueous, catechin-rich heartwood extract of *A. catechu* exhibits potent anticancer activity as demonstrated by the prevention of squamous cell, mammary, and liver cancers in a dose-dependent manner.

ANTI-INFLAMMATORY AND ANALGESIC ACTIVITY

Burnett *et al.* (2007) tested a proprietary mixture of extracts of *A. catechu* and *S. baicalensis* for its ability to inhibit cyclooxygenase and 5-lipoxygenase enzyme activities in *in vitro*, cellular, and *in vivo* models. These two enzymes are important in the production of inflammatory cytokines from arachidonic acid. The results showed that this combination product was able to inhibit these two enzymes and reduce arachidonic acid-induced production of inflammation in a mouse ear-swelling model. No studies were conducted on *A. catechu* alone.

A subsequent study by these investigators (Tseng-Crank *et al.*, 2010) assessed the anti-inflammatory activity of the proprietary *S. baicalensis* and *A. catechu* proprietary blend using lipopolysaccharide as the pro-inflammatory agent in animal and human immortalized cell lines and primary human cells. The combination product had a normalizing effect on the pro-inflammatory genes for cyclooxygenase, tumor necrosis factor, IL-1β, and IL-6 in all cell models. Furthermore, the central controlling factor for these genes, Nuclear factor - kappa B (NF-ƙB), was also downregulated. These results provide a mechanistic understanding of the anti-inflammatory effects of the combination product.

A 90-day oral safety study was conducted on a combination *S. baicalensis* and *A. catechu* product in male and female rats (Yimam *et al.*, 2010). A dose of 1000 mg/kg/day was identified as the no-observed-adverse-effect level, and was the highest dose used in this study. No effects were observed with respect to body weight, feed consumption, clinical observations, organ weights, gross findings, spermatogenesis, estrus staging, ophthalmology, neurology, histopathology, or blood chemistries. These results indicate a very high level of safety.

In another study by Yimam *et al.* (2012), the analgesic activity of a proprietary *S. baicalensis* and *A. catechu* extract combination was assessed in rats and mice using paw edema, formalin test, and abdominal constriction assays. A dose of 100 mg/kg exhibited 58% and 72% inhibition of pain in the abdominal constriction and formalin tests, respectively, while 150 mg/kg reduced pain by about 40% in the paw edema assay. The analgesic effects were believed to be produced through the anti-inflammatory/antioxidant mechanism previously demonstrated.

In a subsequent study, Yimam *et al.* (2013) examined the efficacy of UP446, a combination of baicalin from *S. baicalensis* and (+)-catechin from the heartwood of *A. catechu*, in an experimental rat model of rheumatoid arthritis. A dose of 50 mg/kg of the combination product resulted in significant reductions in pain sensitivity, paw edema, and ankle diameter. With respect to mechanism, the product was shown to decrease the expression of the pro-inflammatory cytokines TNF-α, IL-1β, and IL-6. As previously noted, the combination product is believed to act through the dual inhibition of cyclooxygenase and lipoxygenase.

ANTIMICROBIAL ACTIVITY

Various studies have examined the antimicrobial activity of *A. catechu* extracts, and demonstrated good-to-excellent activity depending on the organism involved. An aqueous extract of *A. catechu* exhibited moderate activity against a multiple drug resistant *Salmonella typhi* (Rani and Khullar, 2004). Patel *et al.* (2009) showed that an aqueous extract of *A. catechu* resin from heartwood exhibited excellent activity against *Bacillus subtilis*, while a petroleum ether extract gave excellent activity against *Pseudomonas aeruginosa*, and a chloroform extract was active against *Staphylococcus aureus.* No identification of active constituents was conducted.

An ethyl acetate extract of heartwood exhibited antimicrobial activity against *B. subtilis, S. aureus, Klebsiella pneumonia,* and *Shigella* species (Joshi *et al.*, 2011). A methanol extract of *A. catechu* was shown to have antimicrobial activities against *B. subtilis, S. aureus, Sal. typhi, Escherichia coli, P. aeruginosa,* and *Candida albicans* (Negi and Dave, 2010). Aqueous and ethanol extracts of *A. catechu* demonstrated moderate activity against hospital isolates of methicillin-resistant *S. aureus* (Voravuthikunchai and Kitpipat, 2005). Lakshmi *et al.* (2011) have also demonstrated that ethanol extracts of A. *catechu* exhibit inhibitory activity against various microbes. Thus, various extracts of *A. catechu* heartwood exhibit antimicrobial activity, but in no case were the active chemical constituents for these activities identified.

DISCUSSION

The aqueous extract of *A. catechu* heartwood has been shown to be a rich source of catechin and epicatechin, with smaller amounts of flavonoids, chemicals with well-recognized antioxidant properties. Extraction with organic solvents as ethyl acetate, ethanol, methanol, hexane, and chloroform results in products with differing chemical compositions and subsequently differing physiological and pharmacological activities. The antioxidant properties of *A. catechu* heartwood extract has been demonstrated in a variety of *in vitro* systems and *in vivo* studies in rats, mice, and cell culture systems. The anti-inflammatory, antineoplastic, and analgesic activities are all believed to be due to the antioxidant activities, although no direct associations have been made.

Various studies involving catechin-rich and epicatechin-rich fractions from other plants have demonstrated antineoplastic (Chen and Zhang, 2007), analgesic (Kuang *et al.*, 2012), and anti-inflammatory properties (Zhong *et al.*, 2012). As a consequence, it is not surprising that various extracts of *A. catechu* exhibit these properties because of the presence of catechins and flavonoids.

Mechanistically, the extracts of *A. catechu* heartwood has been shown to enhance various antioxidant enzymes, increase cellular content of reduced glutathione, which is one of the primary endogenous antioxidants, and inhibit lipid peroxidation and DNA damage. Furthermore, these effects may be mediated by a normalizing effect on the pro-inflammatory genes responsible for the production of inflammatory cytokines, and the downregulation of NF-ĸB, the central controlling factor for these genes (Tseng-Crank *et al.*, 2010).

Most studies involving *A. catechu* heartwood extract have been conducted in rats, mice, and cell culture systems. Monga *et al.* (2011, 2013, 2012) have conducted a series of studies clearly demonstrating the chemoprotective and cancer-preventive activities of the *A. catechu* heartwood extract, and providing much information on the antioxidant activity associated therewith.

Various animal and *in vitro* studies have indirectly demonstrated a lack of adverse events. No animal or human studies have been conducted to specifically address long-term safety. However, extracts of heartwood and other plant parts have been used for hundreds of years without apparent toxicity or adverse effects. *Acacia catechu* heartwood extracts have been used for many years in human medicine as an astringent with no known toxicity. Antioxidant, anti-inflammatory, free radical-scavenging, and tissue-protective effects of heartwood extracts have been well documented, contributing to the overall safety. Furthermore, no adverse events have been reported when *A. catechu* heartwood extracts alone or in combination have been used in human subjects and animals (Burnett *et al.*, 2007; Morgan *et al.*, 2009; Levy *et al.*, 2010, Nadumane, 2011; Yimam *et al.*, 2010, 2012, 2013).

Few human clinical studies have been conducted. The primary studies that have been conducted in humans involve a proprietary extract combination of *S. baicalensis* and *A. catechu* at a dose of 250–500 mg/day for 1–12 weeks, with no adverse effects being reported (Yimam *et al.*, 2010, 2012, 2013). It is not clear what per cent of the extract combination was provided by the *A. catechu.*

Several studies in animals and cell culture systems have also examined the anti-inflammatory and analgesic properties as well as the safety of the proprietary combination of *S. baicalensis* and *A. catechu* extracts (Burnett *et al.*, 2007; Tseng-Crank *et al.*, 2010). Although these results are very promising, the studies do not provide information on *A. catechu* alone and its relative contribution to the proprietary blend.

Finally, as previously noted, *A. catechu* heartwood extract is used as a component of betel quid. Oral cancer and epithelial dysplasia are known to be associated with the chewing of betel quid and are believed to be due to the production of reactive oxygen species and free radicals (Thomas and Kearsley, 1993; Nair *et al.*, 2004; Waris and Nagi, 2014). Because of its antioxidant and free radical-scavenging ability, *A. catechu* heartwood extract may serve as a protectant and antineoplastic in betel quid. *Acacia catechu* extracts have been shown to exhibit antineoplastic and antiproliferative activities (Nadumane, 2011; Ghate *et al.*, 2014).

SUMMARY

Various studies have shown that *A. catechu* heartwood is an excellent source of catechins and epicatechins as well as flavonoids, which have a high degree of antioxidant activity. The antioxidant activity has been well demonstrated by both *in vitro* and *in vivo* studies. The antioxidant activity is believed to be responsible for the anti-inflammatory, antineoplastic, tissue protectant, and analgesic activities that have been demonstrated, and may be related to the antihypertensive and antidiarrheal effects. In spite of the long-term use of *A. catechu* and the general safety of catechins and epicatechins, there is a need for additional well-controlled safety studies in animals and humans. Additional well-controlled human efficacy studies are also needed. Furthermore, few studies have attempted to relate various effects to specific constituents.

Conflict of Interest

The authors have no conflicts of interest to report.

REFERENCES

Arjmandi BH, Ormsbee LT, Elam ML, *et al.*. 2014. A combination of *Scutellaria baicalensis* and *Acacia catechu* extracts for short-term symptomatic relief of joint discomfort associated with osteoarthritis of the knee. *J Med Food* **17**: 707–713.

Bhatia V, Srivastava SP, Srivastava R, *et al.*. 2011. Antihyperglycaemic and aldose reductase inhibitory potential of *Acacia catechu* hard wood and *Tectona grandis* leaves. *Med Chem Res* **20**: 1724–1731.

Bitto A, Squadrito F, Irrera N, *et al.*. 2014. Flavocoxid, a nutraceutical approach to blunt inflammatory conditions. *Med Inflamm* . DOI:10.1155/2014/790851.

Burnett BP, Jia Q, Zhao Y, Levy RM. 2007. A medicinal extract of *Scutellaria baicalensis* and *Acacia catechu* acts as a dual inhibitor of cyclooxygenase and 5-lipoxygenase to reduce inflammation. *J Med Food* **10**: 442–451.

Chaudhari PN, Hatwalne VG. 1970. Effect of *Acacia catechu* on niacin, ascorbic acid and riboflavin status in rats. *J Vitaminol (Kyoto)* **17**: 105–107.

Chaudhari PN, Hatwalne VG. 1971. Effect of *Acacia catechu* on niacin biosynthesis and on niacin requirement in rats. *J Vitaminol (Kyoto)* **17**: 125–127.

Chen L, Zhang HY. 2007. Cancer preventive mechanisms of the green tea polyphenol (−)-epigallocatechin-3-gallate. *Molecules* **12**: 946–957.

Chu NS. 2001. Effects of betel chewing on the central and autonomic nervous systems. *J Biomed Sci* **8**: 229–236.

Ghate NB, Hazra B, Sarkar R, Mandal N. 2014. Heartwood extract of *Acacia catechu* induces apoptosis in human breast carcinoma by altering bax/bcl-2 ratio. *Pharmacogn Mag* **10**: 27–33.

Guleria S, Tiku AK, Singh G, Vyas D, Bhardwaj A. 2011. Antioxidant activity and protective effect against plasmid DNA strand scission of leaf, bark, and heartwood extracts from *Acacia catechu*. *J Food Sci* **76**: C959–C964.

Gunindro N, Devi KP, Singh TI. 2013. Effects of *Acacia catechu* on intestinal absorption of glucose in rats. *J Chem Pharmaceut Res* **5**: 78–81.

Hazra B, Sarkar R, Biswas S, Mandal N. 2010. The antioxidant, iron chelating and DNA protective properties of 70% methanolic extract of "katha" (heartwood extract of *Acacia catechu*). *J Compl Integr Med* **7**: . DOI:10.2202/1553-3840.1335.

Hazra B, Sarkar R, Ghate NB, Chaudhuri D, Mandal N. 2013. Study of the protective effects of Katha (heartwood extract of Acacia catechu) in liver damage induced by iron overload. *J Environ Pathol Toxicol Oncol* **32**: 229–240.

Hiraganahalli BD, Chinampurdur VC, Dethe S, *et al.*. 2012. Hepatoprotective and antioxidant activity of standardized herbal extracts. *Pharmacogn Mag* **8**: 116–123.

Jarald E, Joshi SB, Jain DC. 2009. Biochemical study on the hypoglycemic effects of extract and fraction of *Acacia catechu* Willd in alloxan-induced diabetic rats. *Int J Diabetes Metabol* **17**: 63–69.

Jayasekhar P, Mohanan PV, Rathinam K. 1997. Hepatoprotective activity of ethyl acetate extract of *Acacia catechu*. *Indian J Pharmacol* **29**: 426–428.

Joshi S, Subedi YP, Paudel SK. 2011. Antibacterial and antifungal activity of heartwood of *Acacia catechu* of Nepal. *J Nepal Chem Soc* **27**: 94–99.

Kuang X *et al.* 2012. Effects of intrathecal epigallocatechin gallate, an inhibitor of Toll-like receptor 4, on chronic neuropathic pain in rats. *Eur J Pharmacol* **676**: 51–56.

Kumar U, Mishra M, Prakash V. 2013. Assessment of antioxidant enzymes and free radical scavenging activity of selected medicinal plants. *Free Radicals Antioxid* **2**: 58–63.

Lakshmi T, Geetha RV, Roy A. 2011. *In vitro* evaluation of antibacterial activity of *Acacia catechu* Willd heartwood extract. *Int J Pharm Biosci* **2**: 8188–8192.

Levy RM, Khokhlov A, Kopenkin S, *et al.*. 2010. Efficacy and safety of flavocoxid, a novel therapeutic, compared to naproxen: a randomized multicenter controlled trial in subjects with osteoarthritis of the knee. *Adv Ther* **27**: 731–742.

Li XC, Liu C, Yang LX, Chen RY. 2011. Phenolic compounds from the aqueous extract of *Acacia catechu*. *J Asian Nat Prod Res* **13**: 826–830.

Monga J, Chauhan CS, Sharma M. 2011. Human epithelial carcinoma cytotoxicity and inhibition of DMBA/TPA induced squamous cell carcinoma in Balb/c mice by *Acacia catechu* heartwood. *J Pharm Pharmacol* **63**: 1470–1482.

Monga J, Chauhan CS, Sharma M. 2013. Human breast adenocarcinoma cytotoxicity and modulation of 7,12-dimethylbenz[a]anthracene-induced mammary carcinoma in Balb/c mice by *Acacia catechu* (L.f.) Wild heartwood. *Integr Cancer Ther* 12. 347–362.

Monga J, Chauhan CS, Sharma M. 2012. Chemopreventive efficacy of (+)-catechin-rich aqueous extract of *Acacia catechu* Willd. heartwood against 7,12-dimethylbenz[a]anthracene-induced hepatocarcinoma in Balb/c mice. *J Environ Pathol Toxicol Oncol* **31**: 313–323.

Morgan SL, Baggott JE, Moreland L, Desmond R, Kendrach AC. 2009. The safety of flavocoxid, a medical food, in the dietary management of knee osteoarthritis. *J Med Food* **12**: 1143–1148.

Nadumane VK. 2011. Evaluation of the anticancer and cytotoxic potentials of *Acacia catechu* extracts *in vitro*. *J Nat Pharm* **2**: 190–195.

Naik GH *et al.* 2003. Comparative antioxidant activity of individual herbal components used in Ayurvedic medicine. *Phytochemistry* **63**: 97–104.

Nair U, Bartsch H, Nair J. 2004. Alert for an epidemic of oral cancer due to use of betel liquid substitutes gutha and pan masala: A review of agents and causative mechanisms. *Mutagen* **19**: 251–262.

Negi BS, Dave BP. 2010. *In vitro* antimicrobial activity of *Acacia catechu* and its phytochemical analysis. *Indian J Microbiol* **50**: 369–374.

Noorani AA, Mulla K, Patidar K, Soni M. 2010. Free-radical scavenging activity of ethyl acetate extract of *Acacia catechu* (Willd.) Inventi Rapid: Ethnopharmacol. 2010. http://www.inventi.in/Article/ep/114/10.aspx.

Ojha D, Singh G, Upadhayaya YN. 1969. Clinical evaluation of *Acacia catechu*, Willd. (Khadira) in the treatment of lepromatous leprosy. *Int J Lepr Other Mycobact Dis* **37**: 302–307.

Patel JD, Kumar V, Bhatt SA. 2009. Antimicrobial screening and phytochemical analysis of the resin part of *Acacia catechu*. *Pharmaceut Biol* **47**: 34–37.

Patil S, Jolly CL, Narayanan S. 2003. Free radical scavenging activity of *Acacia catechu* and *Rotula aquatica*: implications in cancer therapy. *Indian Drugs* **40**: 328–332.

Rani P, Khullar N. 2004. Antimicrobial evaluation of some medicinal plants for their anti-enteric potential against multi-drug resistant *Salmonella typhi*. *Phytother Res* **18**: 67–673.

Rahmatullah M, Hossain M, Mahmud A, *et al.*. 2013. Antihyperglycemic and antinociceptive activity evaluation of "khoyer" prepared from boiling the wood of *Acacia catechu* in water. *Afr J Tradit Complement Altern Med* **10**: 1–5.

Ray D, Sharatchandra K, Thokchom IS. 2006. Antipyretic, antidiarrheal, hypoglycemic and hepatoprotective activities of ethyl acetate extract of *Acacia catechu Willd.* in albino rats. *Indian J Pharmacol* **38**: 408–413.

Sham JSK, Chiu KW, Pang PKT. 1984. Hypotensive action of *Acacia catechu*. *Planta Med* **50**: 177–180.

Shen D, Wu Q, Wang M, Yang Y, Lavoie EJ, Simon JE. 2006. Determination of the predominant catechins in *Acacia catechu* by liquid chromatography/electrospray ionization-mass spectrometry. *J Agric Food Chem* **54**: 3219–3224.

Singh KN, Mittal RK, Barthwal KC. 1976. Hypoglycemic activity of *Acacia catechu, Acacia suma*, and *Albizzia odoratissima* seed diets in normal albino rats. *Indian J Med Res* **64**: 754–757.

Sulaiman CT, Balachandran I. 2012. Total phenolics and total flavonoids in selected Indian medicinal plants. *Indian J Pharm Sci* **74**: 258–260.

Sulaiman CT, Gopalakrishnan VK, Balachandran I. 2011. Spectrophotometric determination of antioxidant potential of selected *Acacia* species. *Med Plants-Int J Phytomed Relat Ind* **3**: 289–292.

Thomas S, Kearsley J. 1993. Betel quid and oral cancer: a review. *Oral Oncol Eur J Cancer* **29B**: 251–255.

Tseng-Crank J *et al.* 2010. A medicinal plant extract of *Scutellaria baicalensis* and *Acacia catechu* reduced LPS-stimulated gene expression in immune cells: a comprehensive genomic study using QPCR, ELISA, and microassay. *J Diet Suppl* **7**: 253–272.

Valte V, Singh TI, Singh OJ, Babycha L, Aruna S. 2012. Protective effect of ethyl acetate extract of *Acacia catechu* in carbon tetrachloride induced hepatotoxicity. *Indian Med Gaz* April **12**: 159–162.

Voravuthikunchai SP, Kitpipat L. 2005. Activity of medicinal plant extracts against hospital isolates of methicillin-resistant *Staphylococcus aureus*. *Clin Microbiol Infect* **11**: 510–512.

Waris S, Nagi AH. 2014. Epithelial dysplasia in oral mucosa and its association with betel quid chewing. *J Cytol Histol* **5**: DOI:10.4172/2157-7099.1000258.

Yimam M, Zhao Y, Ma W, Jai Q, Do SG, Shin JH. 2010. 90-Day oral toxicity study of UP446, a combination of defined extracts of *Scutellaria baicalensis* and *Acacia catechu*, in rats. *Food Chem Toxicol* **48**: 1202–1209.

Yimam M, Browell L, Hodges M, Jai Q. 2012. Analgesic effects of a standardized bioflavonoid composition from *Scutellaria baicalensis* and *Acacia catechu*. *J Diet Suppl* **9**: 155–165.

Yimam M, Brownell L, Pantier M, Jia Q. 2013. UP446, analgesic and anti-inflammatory botanical composition. *Pharmacognosy Res* **5**: 139–145.

Zhong Y, Chiou YS, Pan MH, Shahidi F. 2012. Anti-inflammatory activity of lipophilic epigallocatechin gallate (EGCG) derivatives in LPS-stimulated murine macrophages. *Food Chem* **134**: 742–748.

