A review of antioxidant and pharmacological properties of phenolic compounds in *Acacia confusa*

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ABSTRACT

In the present review article, the phytochemical, antioxidant and pharmacological studies are congregated and summarized concerning the current knowledge of the phenolic compounds of a traditional medical plant *Acacia confusa* in Taiwan. This plant is native to Taiwan and South-East Asia. It possesses major pharmacological activities, including antioxidant and radical scavenging activity, hepatoprotective effect, xanthine oxidase inhibition, semicarbazide-sensitive amine oxidase inhibition, angiotensin I converting enzyme inhibition, antihyperuricemic effect and anti-inflammatory activity. Phenolic compounds, especially flavonoids, flavonol glycoside and phenolic acid derivatives, are the main phytochemical compounds isolated from different plant parts of *A. confusa*. Recent interest in this species has focused on pharmacological investigations of the phytochemicals which exhibit potent antioxidant activity based on the multiple phenolic functionalities. The consequence of this review will further extend the potential applications of this plant and offer persuasive support to its future use in the fields of clinical medicine and health functional food.

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1. Introduction

Acacia confusa Merr. (Leguminosae) is an endemic species of Taiwan and is one of the most widespread plants. *A. confusa* is a very adaptable and fast-growing plant suited to living in extreme conditions.[1] It can be found along the coast and river, slightly in from the high tide mark; up to the temperate forests of the higher mountains, but well below the freeze line. However, it usually tends to be more common below 1000 m elevation and temperatures in the range of 20–30 °C. In the wild, it can grow in the soils with poor nutritional value, such as hard clay, silt, dirt and rocky plain and hills. In southern Taiwan, where the winter months can be totally without rain and the summer months can have very heavy rain and typhoons, this plant is able to withstand and grow quite comfortably in a range of climates.[2] *A. confusa*, unlike many plant species of the Leguminosae family, forms a symbiotic association with rhizobia in which its root plays host to them. Many of the associations fix nitrogen from the atmosphere and eventually make it available to the plant and fertile to the surrounding soil.[3]

A. confusa is an evergreen plant (Fig. 1A). The stems and roots are incredibly hard and extremely strong. The sapwood is pale yellow and the heartwood is chocolate brown from the tannins. The barks are rough without ridge and spines. The leaves only appear on seedlings and young plants, and the phyllodes grow on mature plants (Fig. 1B). The phyllodes are dull green, 8–10 cm long and 10–15 mm wide, alternate, narrowly curved-shaped, slightly thickened, hairless, with 3–5 slim parallel veins from the base. The flower clusters of bright yellow balls roughly in 6–20 mm diameter emerge from the twigs (Fig. 1B). In Taiwan, flowering season is usually the coming summer, but, sometimes, it may occur year round. The fruits (pods) are narrow and flat, 5–10 cm long, 8–10 mm wide, dark brown, and split open. The seeds are beanlike, around 5 mm long, 4–8 pre pod, elliptical, dark brown, slightly flattened and shiny.[2]

In Taiwan, *A. confusa* was used as a traditional medicine. The aqueous extract of *A. confusa* leaves was applied to cure wounds and antiblood stasis.[1] The commercial and industrial uses of *A. confusa* were fire wood, charcoal-making, railroad tie, mining construction and mushroom cultivation.[4] For water and soil

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Fig. 1. Plants (A), flowers and phyllodes (B) of *Acacia confusa*.

conservation, this plant is planted in the wilderness for a long time because its root system is extremely strong and can grow extensively and deeply into the ground. Recently, this plant has shown great potential for air pollution prevention because of its remarkable carbon dioxide sequestration ability and foliar dust retention.[5] The bark and wood of *A. confusa*, like many other *Acacia* species, are rich in tannins which are used to dye and stain clothes and tan leather. Due to the high content of tannins and phenolic compounds, many studies have focused on the phytochemistry of *A. confusa* extract in recent years.[6] The aim of this contribution is to review the current literatures on the bioactivities and active compounds of *A. confusa*.

2. Phytochemistry

Secondary metabolite researches have been carried out on *A. confusa* and have led to the isolation of phenolic acid derivatives and flavonoids from its different plant parts. There have been 55 compounds, including 13 phenolic acid derivatives, 3 caffeic acid derivatives, 21 flavonoids, 13 flavonol glycosides, 1 lignan and 4 alkaloids, isolated from the extracts of different plant parts of *A. confusa*. The heartwood extract and the root extract contain flavonoids and phenolic acid derivatives. The bark extract is almost phenolic acid derivatives and caffeic acid derivatives. The extracts of the leaves, branches, twigs, flowers and buds contain flavonol glycosides, some flavonoids and phenolic acid derivatives. The flavonoids in the heartwood and the root extract contain 7,8-dihydroxyflavonoids rather than the usual 5,7-dihydroxyflavonoids in other parts. The following section collected the chemical names of the isolated compounds, and their chemical structures are shown in Fig. 2.

2.1. Constituents in the wood

Ethanol (70%) was used to extract the heartwood. For phenolic acid derivatives, like 3,4-dihydroxybenzoic acid (**3**), 3,4-dihydroxybenzoic acid methyl ester (**4**), 3,4-dihydroxybenzoic acid ethyl ester (**5**) and 3-hydroxy-4-methoxybenzoic acid (**7**) were isolated.[7,8]

Thirteen flavonoids, including 3 flavanols, 1 flavanone, 1 flavanonol, 2 flavones, 5 flavonols and 1 chalcone, were isolated.[7–9] Three flavanols are melacacidin (**18**), 4-*O*-methylmelacacidin (**20**)

and 4′-*O*-methylmelacacidin (**21**). One flavanone is 7,8,3′,4′-tetrahydroxyflavanone (**22**). One flavanonol is *trans*-3,7,8,3′,4′-pentahydroxyflavanone (**24**). Two flavones are 7,3′,4′-trihydroxyflavone (**25**) and 7,8,3′,4′-tetrahydroxyflavone (**26**). Five flavonols are 7,3′,4′-trihydroxy-3-*O*-methylflavonol (**27**), melanoxetin (**28**), transilitin (**29**), 4′-*O*-methylmelanoxetin (**30**) and 4′-*O*-methyltransilitin (**31**). One chalcone is okanin (**32**).

2.2. Constituents of the root

Fourteen compounds, including 1 phenolic acid, 11 flavonoids and 2 alkaloids were isolated from 95% ethanolic root extract. These compounds are 3,4-dihydroxybenzoic acid (**3**), melacacidin (**18**), isomelacacidin (**19**), 4-*O*-methylmelacacidin (**20**), 4′-*O*-methylmelacacidin (**21**), *cis*-3,7,8,3′,4′-pentahydroxyflavanone (**23**), *trans*-3,7,8,3′,4′-pentahydroxyflavanone (**24**), melanoxetin (**28**), transilitin (**29**), okanin (**32**), (+)-catechin (**33**), (-)-epicatechin (**34**), *N*-methyltryptamine (**52**) and *N,N*-dimethyltryptamine (**53**).[10,11] Flavonoids are the main constituents of the heartwood and the root, however, the constituents of the root contain alkaloids not found in heartwood.

2.3. Constituents of the bark

Phenolic acid derivatives are the most abundant constituents in the 70% ethanolic bark extract. Fifteen phenolic acid derivatives, including 4-hydroxybenzoic acid (**1**), 4-hydroxybenzoic acid ethyl ester (**2**), 3,4-dihydroxybenzoic acid (**3**), 3,4-dihydroxybenzoic acid methyl ester (**4**), 3,4-dihydroxybenzoic acid ethyl ester (**5**), 3,4-dihydroxybenzoic acid butyl ester (**6**), 3-hydroxy-4-methoxybenzoic acid (**7**), 4-hydroxy-3-methoxybenzoic acid (**8**), 4-hydroxy-3,5-dimethoxybenzoic acid (**9**), 4-hydroxy-3,5-dimethoxybenzoic acid ethyl ester (**10**), gallic acid (**11**), gallic acid ethyl ester (**13**), 3,4-dihydroxy-*trans*-cinnamic acid (**14**), 3,4-dihydroxy-*trans*-cinnamic acid ethyl ester (**15**), 3,4-dihydroxy-*trans*-cinnamic acid pentyl ester (**16**) and one lignan, (-)-lyoniresinol (**17**), were isolated.[12,13]

The bark of *A. confusa* is a good source of condensed tannins (proanthocyanidins). The stem bark extract and the root bark extract comprised 247.76 ± 10.93 and 280.70 ± 11.75 mg/g of extractable condensed tannins. According to the results of MALDI-TOF MS, the degree of polymerization (DP) for both extracts can
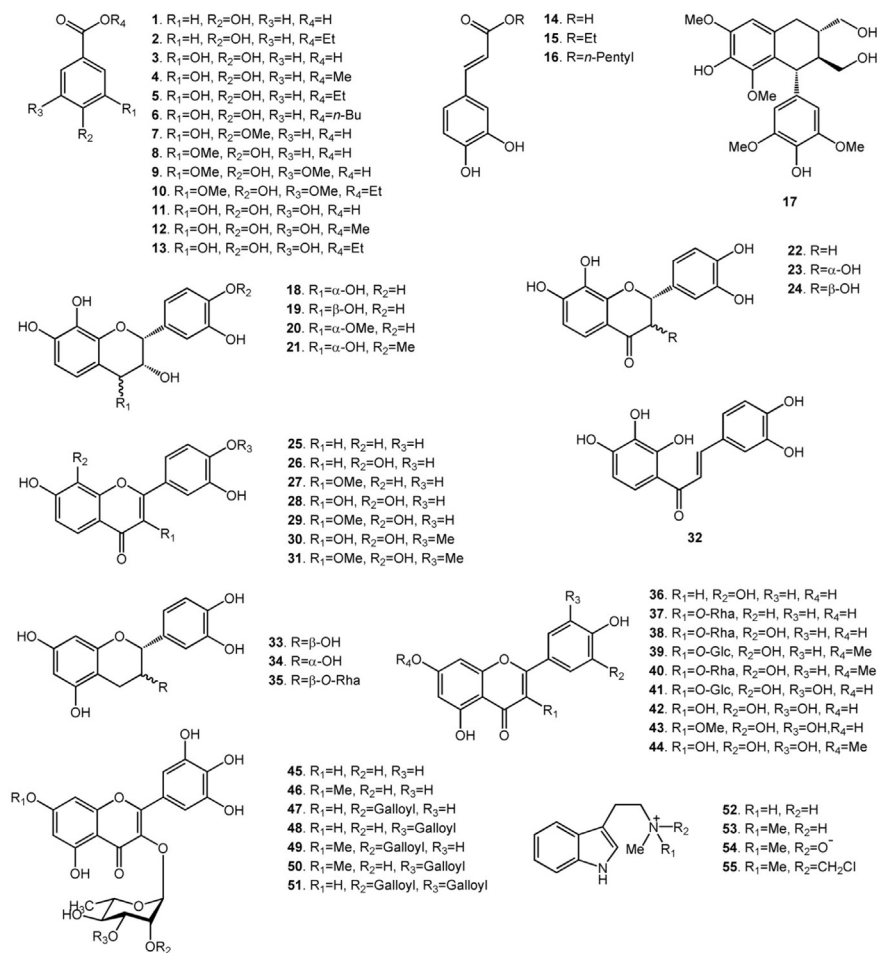
Fig. 2. Compounds isolated from *Acacia confusa*.

reach 12 and 11, respectively, and the structures of the identified condensed tannins show almost B-type bonding.[6]

2.4. Constituents of the branches and twigs

The branches and the twigs were extracted respectively in 95% ethanol. (+)-Catechin (**33**), (-)-epicatechin (**34**), catechin-3-O-α-rhamnopyranoside (**35**) and quercitrin (quercetin-3-O-α-rhamnopyranoside) (**38**) were isolated from the branches extract and luteolin (**36**), isomyricetin (myricetin-3-O-β-glucopyranoside) (**41**), myricitrin (myricetin-3-O-α-rhamnopyranoside) (**45**) and myricetin-3-O-(2″-O-galloyl)-α-rhamnopyranoside (**47**) were isolated from the twigs.[14] Chemical composition analysis revealed the total flavonoids content (TFC) is in the order of twigs (7.7 ± 0.3 mg of quercetin equivalent (QE)/g), branches (2.1 ± 0.0 mg of QE/g) and branch bark (0.9 ± 0.0 mg of QE/g). On the contrary, the total proanthocyanidins content (TPAC) is in the order of the branch bark (128.4 ± 7.6 mg of catechin equivalent (CE)/g), branches (30.4 ± 0.4 mg of CE/g) and twigs (non-detected). Hence, the chemical composition analysis is inconsistent with the compounds isolated from the branch and twig extracts. According to the flavonoids synthesis mechanisms, flavones and flavonols are the upstream products and flavanols are the down-stream products.[15] Since flavanols are the monomers of condensed tannins, it is postulated when the twigs gradually grow into branches, the condensed tannin content is also augmented. Condensed tannins have been closely associated with plant defense mechanisms

against mammalian herbivores, birds, insects and fungi.[16–18] Therefore, variations of flavonoids in different plant parts of *A. confusa* may be involved in plant protective effects, which agrees with previous reports.[19,20]

According to recent studies, the constituents of the branches and twigs are 5,7-dihydroxyl flavonoids which have the same flavonoids synthesis mechanism as reported in the literature.[15] When the stem keeps growing and the heartwood part is formed, 5,7-dihydroxyl flavonoids are transformed to 7,8-dihydroxyl flavonoids and its sugar moiety is removed. Last, 7,8-dihydroxyl flavonoids are accumulated in the heartwood part.

2.5. Constituents of the leaves

Methanol and hot water were used to extract the leaves of *A. confusa*. Flavonol glycosides, like quercitrin (**38**), isomyricetin (**41**), myricitrin (**45**), 7-O-methylmyricitrin (myricetin-3-O-α-rhamnopyranoside-7-O-methyl ether) (**46**), myricetin-3-O-(2″-O-galloyl)-α-rhamnopyranoside (**47**), myricetin-3-O-(3″-O-galloyl)-α-rhamnopyranoside (**48**), myricetin-3-O-(2″-O-galloyl)-α-rhamnopyranoside-7-methyl ether (**49**), myricetin-3-O-(3″-O-galloyl)-α-rhamnopyranoside-7-methyl ether (**50**) and myricetin-3-O-(2″,3″-di-O-galloyl)-α-rhamnopyranoside (**51**), are major constituents of the leaf extracts.[21–23] Besides, flavonoids and phenolic acid derivatives, like (+)-catechin (**33**), (-)-epicatechin (**34**), luteolin (**36**), myricetin (**42**), 3-O-methylmyricetin (**43**), europetin (7-O-methylmyricetin) (**44**), gallic acid (**11**) and gallic acid methyl ester

(**12**) were also found in the leaf extracts.[21,22] The leaf extracts also contained condensed tannins and the content of the extractable condensed tannins was 64.17 ± 1.44 mg/g.[6]

2.6. Constituents of the flowers and buds

Gallic acid (**11**), afzelin (kaempferol-3-O-α-rhamnopyranoside) (**37**), quercitrin (**38**), rhamnetin-3-O-α-glucopyranoside (quercetin-3-O-α-glucopyranoside-7-O-methyl ether) (**39**), rhamnitrin (quercetin-3-O-α-rhamnopyranoside-7-O-methyl ether) (**40**), myricitrin (**45**), 7-O-methylmyricitrin (**46**) were found to be the major secondary metabolites in the ethanolic extract of *A. confusa* flowers and buds.[24,25]

2.7. Alkaloids of the shrubs

The alkaloids study on the methanolic extract of *A. confusa* shrubs has led to the isolation of four tryptamine compounds, and they are N-methyltryptamine (**52**), N,N-dimethyltryptamine (**53**), N,N-dimethyltryptamine-N-oxide (**54**) and N-chloromethyl-N,N-dimethyltryptamine (**55**).[26]

3. Biological activities

The biological activities of the extractives from *A. confusa* have been investigated and found that they possess various activities in antioxidant, scavenging radical, anti-inflammatory, anti-virus, inducing hallucination, etc (Fig. 3 and Table 1). The following section introduces their effective constituents and related bioactivities.

3.1. Antioxidant and radical scavenging activities

Evaluation of the antioxidant potency of the extracts from different plant parts using Folin-Ciocalteu's method shows the root extract has the highest total phenolic contents (TPC) (652.2 mg gallic acid equivalence (GAE)/g),[10] followed by the heartwood extract (529.7 mg GAE/g),[27] the bark extract (470.6 mg GAE/g),[27] the branches extract (348.2 mg GAE/g),[14] the leaves extract (190.2 mg GAE/g),[28] the buds extract (173.3 mg GAE/g),[24] the twigs extract (121.4 mg GAE/g)[14] and the flowers extract (105.1 mg GAE/g).[24] The isolated compounds show strong antioxidant activity and radical scavenging activity *in vitro* mainly because their phenolic groups form catechol group and pyrogallol group in their structures. Flavonoids isolated from the heartwood and the root extracts have unique 7,8-dihydroxyl structures which lead to an increment in their antioxidant activity. Further, okanin (**32**) and melanoxetin (**28**) are the strongest antioxidant flavonoids which show the

lowest IC_{50} values of radical scavenging ability against DPPH (2,2-diphenyl-1-picrylhydrazyl) (3.1 and 3.1 μM) and superoxide (2.2 and 2.5 μM) radicals and highest trolox equivalent antioxidant capacity (TEAC, 5.4 and 4.4 mmol of Trolox equivalence (TE)/mmol) and reducing power (5.3 and 5.0 mmol of TE/mmol).[10] Gallic acid (**11**) found in bark, leaves, flowers and buds extracts shows remarkable DPPH and superoxide radical scavenging activity, with IC_{50} values of 8.2 and 12.4 μM, and TEAC of 5.2 mmol of TE/mmol, which is the best antioxidant for phenolic acid derivatives.[12,13] Myricetin-3-O-(2″-O-galloyl)-α-rhamnopyranoside (**47**) isolated from the twigs and the leaves extracts is the most active flavonol glycoside, exhibiting IC_{50} values of 3.9 and 8.7 μM for inhibiting DPPH and superoxide radicals and TEAC of 6.9 TE/mmol.[22] Therefore, extracts from different plant parts of *A. confusa* contain abundant amounts and various types of phenolic compounds and possess excellent antioxidant activity *in vitro*. The *in vivo* antioxidant efficacies of the extract in *A. confusa* twig had been preliminarily evaluated by the model organism, *Caenorhabditis elegans*.[29] Since twig extract could effectively remove the reactive oxygen species (ROS) in *C. elegans*, the oxidative stress resistance of *C. elegans* was enhanced following pretreatment with the extract.

ROS has harmful effects on DNA damage, lipid peroxidation and oxidative deactivation of specific enzymes and proteins on the cells, which have been implicated in the pathogenesis of many human diseases, such as Alzheimer's disease, amyotrophic, anxiety, atherosclerosis, asthma, cancer, degenerative eye diseases, depression, diabetes, epilepsy, Huntington's disease lateral sclerosis, inflammatory joint disease, multiple sclerosis, Niemann-pick diseases, Parkinson's disease, schizophrenia, senile dementia, etc.[30,31] Use of natural phenolic compounds and flavonoids as an antioxidant has been advised as a common treatment to deal with these disorders. Accordingly, the antioxidant potential of different plant parts of *A. confusa* and their isolated compounds can be further discovered in ameliorating the oxidative stress related disorders.

3.2. Hepatoprotective effects

The ethyl acetate fraction of *A. confusa* bark extract and its active compounds, gallic acid (**11**), demonstrated excellent protective effect against carbon tetrachloride (CCl_4)-induced chronic liver injury in rats.[32] The pathological histology result for the rats liver showed dietary supplementation with gallic acid (**11**) at a dose of 50 mg/kg inhibited big vacuole formation and reduced small vacuole formation by 78%, and decreased inflammation by 57%. It can significantly reduce the plasma levels of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) by 94% and 100%, respectively, which have been considered effective indicators of hepatic injury.
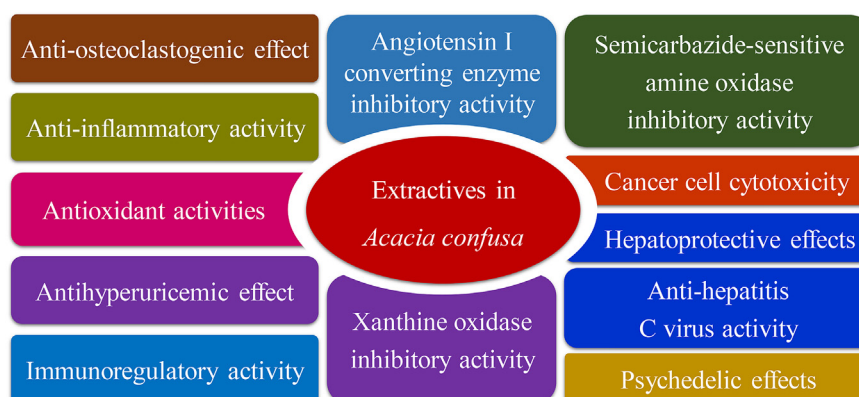

Fig. 3. Summary of the main medical properties of *Acacia confusa*.

Table 1
Medical benefits and pharmacologic properties of the major constituents in *Acacia confusa*.

Pharmacologic properties	Major constituents[a]	References
Antioxidant and radical scavenging activities	**11**, **28**, **32**, **47**	10, 12, 13, 22
Hepatoprotective effects	**11**	32
Xanthine oxidase inhibitory activity	**26**, **28**, **32**, **36**	9, 14, 41
Semicarbazide-sensitive amine oxidase inhibitory activity	**47**, **48**, **49**, **50**, **51**	47
Angiotensin I converting enzyme inhibitory activity	**49**, **50**, **51**	47
Antihyperuricemic effect	**18**, **21**, **28**, **29**, **32**	9
Anti-inflammatory activity	**28**	7
Anti-hepatitis C virus activity	Ceramides	57
Immunoregulatory activity	**28**, **47**[b]	68, 73[b]
Anti-osteoclastogenic effect	**25**[b], **26**[b]	74[b] 75[b]
Cancer cell cytotoxicity	**47**, **48**, **49**, **50**	23
Psychedelic effects	**53**[b]	79[b] 80[b] 81[b] 82[b] 83[b] 84[b] 85[b] 86[b]

[a] The chemical structures of the major constituents are referred to Fig. 2.
[b] Pharmacologic properties of the constituents were determined from other plant species or commercial suppliers.

Gallic acid (**11**) can elevate the activities of antioxidant enzymes, such as superoxide dismutase (SOD), glutathione reductase (GRD), glutathione peroxidase (GPX) and catalase (CAT). For the erythrocytes, the activities of SOD, GPX, CAT and GRD strongly increased from 19, 2.3 and 852 U/mg and 2.1 U/g to 97, 4.9 and 1939 U/mg and 5.1 U/g following treatment with gallic acid (**11**). For liver tissues, the activities of GPX, CAT and GRD increased from 0.21, 48 and 0.49 U/g to 0.32, 79 and 0.54 U/g, and the activities of SOD decreased from 6.9 U/g to 5.6 U/g. Gallic acid (**11**) treatments can obviously reduce the lipid peroxidation level and oxidative stress in plasma and also in liver tissues. By using the thiobarbituric acid reactive substances (TBARS) method, the lipid peroxidation level decreased from 6.2 μM to 3.3 μM and 57 nmol/g liver to 39 nmol/g liver for the plasma and the liver tissues, respectively. The oxidative stress was defined as a reciprocal of the ratio of glutathione (GSH) and oxidized glutathione (GSSG). The GSH/GSSG ratio strongly increased from 27 to 107 and 15 to 93 for the erythrocytes and the liver tissues. Molecular biology study illustrated the expression of hepatic CYP2E1, the major isozyme involved in CCl_4 bioactivation and subsequent free radical production, was significantly decreased by 45% by gallic acid (**11**). In sum, the hepatoprotective effects of gallic acid (**11**) may be due to regulating the activities of antioxidant enzymes and suppressing lipid peroxidation and CYP2E1 activation.

According to the World Health Organization, chronic liver diseases derived from alcoholic liver disease, fatty liver disease and especially viral hepatitis, etc., remain one of the major threats to public health and are a worldwide problem, causing up to 1.45 million deaths worldwide annually.[33,34] Although modem medicine provides great advances in liver disease treatment, no effective drug is available that protects the liver from damage, stimulates liver function or helps regenerate hepatic cells.[35,36] Phytochemicals from traditional medical plants have been evaluated for their hepatoprotective effects involved with free radical scavenging activities, antioxidant properties and adoptogenic effects.[37–39] The treatment of liver diseases using natural remedies and their derivatives has a long history and are still used all over the world. Liver protective plants have all manner of chemical constituents, like phenolic compounds, flavonoids, coumarins, monoterpenes, glycosides, alkaloids and xanthenes.[40] Tung et al.[32] reported the hepatoprotective effects of gallic acid (**11**) isolated from *A. confusa* bark extract show exciting progress in the discovery of effective liver protective agents, especially at present, with the urgent need for innovative drugs.

3.3. Xanthine oxidase (XOD) inhibitory activity

A. confusa heartwood extract exhibited remarkable inhibitory activity against XOD *in vitro*. Tung and Chang[41] reported okanin (**32**) showed the strongest XOD inhibitory effect with an IC_{50} value of 0.076 μM, followed by melanoxetin (**28**, 0.274 μM), allopurinol (positive control, 4.784 μM), and 7,8,3′,4′-tetrahydroxyflavone (**26**, 10.488 μM). The results of the kinetics and molecular mechanisms demonstrated melanoxetin (**28**) and 7,8,3′,4′-tetrahydroxyflavone (**26**) were in a competitive mode, as well as allopurinol, and okanin (**32**) was in a noncompetitive mode. Further studies[41,42] demonstrated melanoxetin (**28**) is a better XOD inhibitor than the commercial drug, allopurinol for two reasons. First, at the same dosage of inhibitor, melanoxetin (**28**) showed lower heat release than allopurinol in exothermic XOD and xanthine reaction. Second, the Michaelis constants (K_m) of XOD and xanthine reaction were 34.6 and 24.5 μM for melanoxetin (**28**) and allopurinol, respectively.[9] The molecular docking studies showed the melanoxetin (**28**) molecule occupies the same binding site as allopurinol, and its carbonyl and multihydroxyl functions may contribute to a higher binding affinity to XOD than allopurinol.[9] Besides, luteolin (**36**) isolated from the twigs extract showed excellent XOD inhibitory activity with an IC_{50} value of 11.6 μM.[14]

XOD plays an important role in the catabolic sequence of the purine nucleotide metabolism in some species, including humans.[42] It not only catalyzes the serial oxidation processes in the transformation of xanthine from hypoxanthine and further from xanthine to uric acid, but also generates ROS, like hydrogen peroxide and superoxide anion radicals, accompanying the catalyzed reaction. Consequently, XOD inhibitors are employed to interrupt the synthesis of uric acid in the final step, decreasing the ROS level in the human body, and promoting the production of anti-inflammatory agents to relieve the symptoms from the disease.[43–46] It is surprising okanin (**32**) and melanoxetin (**28**) exhibit much better XOD inhibitory activity than allopurinol, which is one of the clinical drugs for treating hyperuricemia and acute gout, and shows great potential in the development of new drugs.

3.4. Semicarbazide-sensitive amine oxidase (SSAO) inhibitory activity

Lee et al.[47] reported five myricetin galloglycosides were isolated from the leaves of *A. confusa* and showed inhibitory activity against SSAO (EC 1.4.3.6). Among them, myricetin-3-O-(3″-O-galloyl)-α-rhamnopyranoside-7-methyl ether (**50**) and myricetin-3-O-(2″,3″-di-O-galloyl)-α-rhamnopyranoside (**51**) showed the highest inhibitory activity with IC_{50} values of 36.16 μM and 39.35 μM, respectively, followed by myricetin-3-O-(2″-O-galloyl)-α-rhamnopyranoside-7-methyl ether (**49**), myricetin-3-O-(3″-O-galloyl)-α-rhamnopyranoside (**48**), myricetin-3-O-(2″-O-galloyl)-α-rhamnopyranoside (**47**).

Lee et al.[47] also found these five compounds showed an identical order for inhibitory activity against SSAO and DPPH radical scavenging activity. SSAO is present in plants, microorganisms, and in many organs of mammals.[48] It converts primary amines into the corresponding aldehydes, generating hydrogen peroxide and ammonia. It has been proven plasma SSAO is increased in diabetes mellitus and heart failure and SSAO is implicated in atherosclerosis, endothelial damage, and glucose transport into adipocytes. Therefore, myricetin galloglycosides isolated from the leaf extract of *A. confusa* show benefits to human health.

3.5. Angiotensin I converting enzyme (ACE) inhibitory activity

Myricetin-3-O-(2″,3″-di-O-galloyl)-α-rhamnopyranoside (51), myricetin-3-O-(3″-O-galloyl)-α-rhamnopyranoside-7-methyl ether (50) and myricetin-3-O-(2″-O-galloyl)-α-rhamnopyranoside-7-methyl ether (49) isolated from the leaf extract of *A. confusa* showed inhibitory activities against ACE (EC 3.4.15.1), with IC_{50} values of 19.82, 60.32 and 151.90 μM.[47] Lee et al.[47] suggested ACE inhibitory activity is referring to the gallic acid groups. ACE, a dipeptide-liberating exopeptidase, is involved in the blood pressure regulation around the renin-angiotensin system.[49] ACE inhibitor is the preferred class of antihypertensive drugs, due to its low adverse side-effects.[50]

3.6. Antihyperuricemic effect

Tung et al.[9] reported *A. confusa* heartwood extract can significantly suppress serum uric acid levels in oxonate-induced mice, and lead to the isolation of five active compounds. At an equimolar dose (100 μmol/kg), animals treated with melacacidin (18), 4′-O-methylmelacacidin (21), melanoxetin (28), transilitin (29), okanin (32) and allopurinol (positive control) showed significant reductions in uric acid to 66, 72, 75, 65, 69 and 79%, respectively, relative to the untreated group. Hyperuricemia may lead to the pathogenesis of many serious complications, including: gouty arthritis, gout, stroke, ischemic heart disease, kidney dysfunction, uremia, urolithiasis, etc. Wang et al.[51] reported cinnamaldehyde, the major compound of *Cinnamomum osmophloeum* leaves, significantly reduced the serum uric acid level by 60% at a dosage of 150 mg/kg (1135 μmol/kg). Zhu et al.[52] also found treating with quercetin and rutin at a dosage of 150 mg/kg (496 μmol/kg for quercetin and 245 μmol/kg for rutin) markedly reduced the serum uric acid levels by 36 and 32%, respectively. Therefore, the study of Tung et al.[9] indicates the heartwood extract of *A. confusa* and its isolated five flavonoids as a potential candidate for reducing serum uric acid levels and gout treatment.

3.7. Anti-inflammatory activity

A. confusa heartwood extract showed moderate anti-inflammatory activity in the inhibition of nitric oxide (NO) generation in lipopolysaccharide (LPS)-stimulated RAW 264.7 cells.[7] Melanoxetin (28) is the most active compound which strongly inhibited NO production with an IC_{50} value of 6.9 μM and reduced PGE_2 accumulation by 60% at a dosage of 100 μM. In addition, at a dosage of 50 μM, melanoxetin (28) completely suppressed the iNOS mRNA expression and reduced the cyclooxygenase-2 (COX-2) mRNA expression by 76%. New anti-inflammatory agent discoveries alleviating the symptoms of acute inflammation always attract the attention of pharmacological scientists because they aim to prevent the adverse effect of clinical drugs. On the other hand, chronic inflammation has been gradually proven to be a possible cause of cardiovascular disease, Alzheimer's disease, cancer, allergies, autoimmune diseases and metabolic disorders.[53–56] *A. confusa*

heartwood extract contains abundant amounts of phenolic compounds and flavonoids and shows anti-inflammatory activities, thus, it may have the potential to become a natural source of anti-inflammatory food supplements and drugs.

3.8. Anti-hepatitis C virus activity

Lee et al.[57] showed the glycosyl lipidoids, ceramides, in *A. confusa* stems have inhibition efficacy on anti-hepatitis C virus (HCV). HCV is an enveloped, positive-sense and single-stranded RNA virus belonging to the family *Flaviviridae* that causes chronic hepatitis, cirrhosis and hepatocellular carcinoma in humans.[58,59] The only treatment to cure for HCV infection is using pegylated interferon-α in combination with the nucleoside analog ribavirin which has unfavorable side-effects such as flu-like symptoms, hemolytic anemia and depression.[60] Virus-induced hepatic diseases cause chronic inflammation or proliferation of hepatoma cells involving activation of nuclear factor-kappaB (NF-κB).[61] HCV produces at least 10 individual proteins, 4 structural proteins (core, E1, E2 and p7) and 6 nonstructural proteins (NS2, NS3, NS4A, NS4B, NS5A and NAS5B), and some of them (core, E2, NS3, NS5A) induce hepatocellular carcinoma via promotion of NF-κB mediated through COX-2 expression.[57,62–65] Lee et al.[57] demonstrated the effective constituents in the stems of *A. confusa* can inhibit the HCV RNA replication by suppressing COX-2 expression and NF-κB activation and providing antiviral synergy in combination with INF-α.

3.9. Immunoregulatory activity

Dendritic cells (DCs), one type of professional antigen-presenting cells, present in lymphoid and nonlymphoid tissues, are responsible for regulating immune responses.[66,67] It has been demonstrated the activation of DCs leads to the maturation and expression of pro-inflammatory cytokines such as TNF-α, IL-6 and IL-12.[68–70] Ho et al.[68] reported melanoxetin (28), the major constituent in *A. confusa* heartwood extract, can effectively enhance immune regulation by inhibiting the production of pro-inflammatory cytokines in LPS-stimulated dendritic cells (DCs) at a concentration of 12.5 μM. Besides, LPS also generates ROS which can activate DCs and make immature DCs become mature.[71] Ho et al.[68] found LPS-induced DCs maturation was inhibited by treatment with melanoxetin. Since melanoxetin is a good antioxidant, it is believed that the melanoxetin can enhance immune-regulation by suppressing LPS-generated ROS. Besides, human peripheral blood mononuclear cells (PBMCs) are the important hinge of the immune responses.[72] The activation and proliferation of PBMCs would promote the immune responses.[72] Kuo et al.[73] reported myricetin-3-O-(2″-O-galloyl)-α-rhamnopyranoside (47) has an antiproliferative effect on PBMCs, with an IC_{50} value of 11.9 μM.

3.10. Anti-osteoclastogenic effect

Kang et al.[74,75] demonstrated 7,3′,4′-trihydroxyflavone (25) and 7,8,3′,4′-tetrahydroxyflavone (26) have the potential to treat bone-lytic diseases owing to their anti-osteoclastogenic effect. Osteoclastogenesis causes the immature bone-resorbing osteoclast to mature which is activated by the essential key cytokines, macrophage colony stimulating factor and receptor activator of NF-κB ligand (RANKL).[76,77] Osteoclast is accountable for the primary bone remodeling process via resorption and resorbing of the bone; however, the over activation of osteoclasts would induce bone-related diseases such as postmenopausal osteoporosis, inflammatory arthritis, osteolytic bone metastasis and Paget's bone disease.[78] Kang et al.[74,75] showed 7,3′,4′-trihydroxyflavone (25) and 7,8,3′,4′-tetrahydroxyflavone (26) can inhibit RANKL-induced osteoclast

differentiation by reducing both the expression levels of nuclear factor of activated T cells c1 (a key transcription factor of osteoclast differentiation) and the mRNA of osteoclast marker genes.

3.11. Cancer cell cytotoxicity

Brine shrimp *(Artemia salina)* lethality assay is a rapid, inexpensive, in-house and general bioassay and is usually used for evaluating the cancer cell cytotoxicity of organic compounds, especially for natural products. Lee et al.[23] reported flavonol galloglycosides were the major compounds for *A. confusa* leaf extracts and their anti-hatch activity against brine shrimp were evaluated. Myricetin-3-*O*-(2″-*O*-galloyl)-α-rhamnopyranoside (**47**), myricetin-3-*O*-(3″-*O*-galloyl)-α-rhamnopyranoside (**48**), myricetin-3-*O*-(2″-*O*-galloyl)-α-rhamnopyranoside-7-methyl ether (**49**), myricetin-3-*O*-(3″-*O*-galloyl)-α-rhamnopyranoside-7-methyl ether (**50**) exhibited moderate brine shrimp lethality, with IC$_{50}$ values of 75, 64, 89 and 50 μg/mL.

3.12. Psychedelic effects

It is well known ingesting *N,N*-dimethyltryptamine (**53**) at high concentration causes transient and intermittently visual hallucinations.[79–82] Carbonaro and Gatch[83] reviewed the psychedelic pharmacological mechanisms of *N,N*-dimethyltryptamine (**53**) involved in the interactions of various receptors (i.e. serotonin receptors, trace amine-associated receptors, sigma-1 receptor and ionotropic and metabotropic glutamate receptors) and neurotransmitters (dopamine, acetylcholine and glutamate) in brain, concurrently, *N,N*-dimethyltryptamine (**53**) can be a model of psychiatric disorders in investigations of schizophrenia, depression and anxiety. Besides, since the use of *N,N*-dimethyltryptamine (**53**) is not as harmful as other synthetic hallucinogenic compounds, such as 25I-NBOMe, *N,N*-dimethyltryptamine (**53**) is also considered a powerful media for self-discovery and understanding consciousness.[83–86]

4. Conclusions

Traditional medical plants are potent sources in the drug discovery process and the development of health functional food because their active phenolic compounds are in charge of various antioxidant and pharmacological activities. *A. confusa* is a medical plant endemic in Taiwan and is one of the most widespread plants. To date, this report is the most comprehensive review of the phytochemistry, antioxidant and pharmacological properties of *A. confusa*. Flavonoids, flavonol glycosides and phenolic acid derivatives are the major phytochemical constituents isolated from different plant parts which contain multiple phenolic functionalities exhibiting impressive antioxidant ability. Further, these compounds exhibited remarkable inhibitory activities against XOD, AASO and ACE. Knowledge of the chemical constituents of *A. confusa* is necessary, not only for the discovery of new medical agents, but because such information may be valuable to those interested in the actual worth of folklore drugs. Only a few studies have been done on the biological activities and plausible functional food and medicinal applications of these compounds. Therefore, extensive investigation and development work is needed to exploit their therapeutic utility against diseases and the feasibility of being healthy food supplements.

Conflicts of interest

All authors declare no conflicts of interest.

References

1. Kan WS. Leguminosae. In: Kan WS, ed. *Manual of Medicinal Plants in Taiwan*. vol. 2. Taipei: National Research Institute of Chinese Medicine; 1978:239–240.
2. Huang TC, Ohashi H. Leguminosae. In: Huang TC, ed. *Flora of Taiwan*. second ed. vol. 3. Taipei: National Taiwan University; 1994:160–162.
3. Chen YSG, Wong SM, Whitton BA. Effects of landfill leachate on growth and nitrogen fixation of two Leguminous trees (*Acacia confusa, Leucaena leucocephala*). *Water Air Soil Pollut*. 1999;111:29–40.
4. Wang DM, Wu SH, Su CH, Peng JT, Shih YH, Chen LC. *Ganoderma multipileum*, the correct name for '*G. lucidum*' in tropical Asia. *Bot Stud*. 2009;50:451–458.
5. Wang YC. Carbon sequestration and foliar dust retention by woody plants in the greenbelts along two major Taiwan highways. *Ann Appl Biol*. 2011;159:244–251.
6. Wei SD, Zhou HC, Lin YM, Liao MM, Chai WM. MALDI-TOF MS analysis of condensed tannins with potent antioxidant activity from the leaf, stem bark and root bark of *Acacia confusa*. *Molecules*. 2010;15:4369–4381.
7. Wu JH, Tung YT, Chien SC, et al. Effect of phytocompounds from the heartwood of *Acacia confusa* on inflammatory mediator production. *J Agric Food Chem*. 2008;56:1567–1573.
8. Wu JH, Tung YT, Wang SY, Shyur LF, Kuo YH, Chang ST. Phenolic antioxidants from the heartwood of *Acacia confusa*. *J Agric Food Chem*. 2005;53:5917–5921.
9. Tung YT, Hsu CA, Chen CS, Yang SC, Huang CC, Chang ST. Phytochemicals from *Acacia confusa* heartwood extracts reduce serum uric acid levels in oxonate-induced mice: their potential use as xanthine oxidase inhibitors. *J Agric Food Chem*. 2010;58:9936–9941.
10. Lin HY, Chang ST. Antioxidant potency of phenolic phytochemicals from the root extract of *Acacia confusa*. *Ind Crop Prod*. 2013;49:871–878.
11. Lee TH, Chou CH. Flavonoid aglycones and indole alkaloids from the roots of *Acacia confusa*. *J Chin Chem Soc*. 2000;47:1287–1290.
12. Tung YT, Wu JH, Huang CY, Kuo YH, Chang ST. Antioxidant activities and phytochemical characteristics of extracts from *Acacia confusa* bark. *Bioresour Technol*. 2009;100:509–514.
13. Tung YT, Wu JH, Kuo YH, Chang ST. Antioxidant activities of natural phenolic compounds from *Acacia confusa* bark. *Bioresour Technol*. 2007;98:1120–1123.
14. Hsieh CY, Chang ST. Antioxidant activities and xanthine oxidase inhibitory effects of phenolic phytochemicals from *Acacia confusa* twigs and branches. *J Agric Food Chem*. 2010;58:1578–1583.
15. Dewick PM. *Medicinal Natural Products: a Biosynthetic Approach*. 3th ed. New York: John Wiley & Sons Ltd; 2009.
16. Hassanpour S, Sadaghian M, Maheri-Sis N, Eshratkhah B, Chaichi-Semsari M. Effect of condensed tannin on controlling faecal protein excretion in nematode-infected sheep: *in vivo* study. *J Am Sci*. 2011;7:896–900.
17. Zucker WV. Tannins: does structure determine function? An ecological perspective. *Am Nat*. 1983;121:335–365.
18. Hagerman AE, Buttler LG. The specificity of proanthocyanidin-protein interactions. *J Biol Chem*. 1981;256:4494–4497.
19. Hassanpour S, Maheri-Sis N, Eshratkhah B, Mehmandar FB. Plants and secondary metabolites (tannins): a review. *Int J For Soil Eros (IJFSE)*. 2011;1:47–53.
20. Haslam E. *Plant Polyphenols Vegetable Tannins Revisited*. Cambridge: Cambridge University Press; 1989.
21. Tung YT, Wu JH, Hsieh CY, Chen PS, Chang ST. Free radical-scavenging phytochemicals of hot water extracts of *Acacia confusa* leaves detected by an online screening method. *Food Chem*. 2009;115:1019–1024.
22. Lin SS, Shiau IL, Chang ST. Antioxidant activity of constituents from the methanolic extract of *Acacia confusa* leaves. *Taiwan J For Sci*. 2009;24:61–68.
23. Lee TH, Qiu F, Waller GR, Chou CH. Three new flavonol galloylglycosides from leaves of *Acacia confusa*. *J Nat Prod*. 2000;63:710–712.
24. Tung YT, Chang WC, Chen PS, Chang TC, Chang ST. Ultrasound-assisted extraction of phenolic antioxidants from *Acacia confusa* flowers and buds. *J Separ Sci*. 2011;34:844–851.
25. Wu JH, Huang CY, Tung YT, Chang ST. Online RP-HPLC-DPPH screening method for detection of radical-scavenging phytochemicals from flowers of *Acacia confusa*. *J Agric Food Chem*. 2008;56:328–332.
26. Buchanan MS, Carroll AR, Pass D, Quinn RJ. NMR spectral assignments of a new chlorotryptamine alkaloid and its analogues from *Acacia confusa*. *Magn Reson Chem*. 2007;45:359–361.
27. Chang ST, Wu JH, Wang SY, Kang PL, Yang NS, Shyur LF. Antioxidant activity of extracts from *Acacia confusa* bark and heartwood. *J Agric Food Chem*. 2001;49:3420–3424.
28. Ho ST, Tung YT, Chen YL, Zhao YY, Chung MJ, Wu JH. Antioxidant activities and phytochemical study of leaf extracts from 18 indigenous tree species in Taiwan. *Evid-based Complement Altern Med.*. 2012, 215959 https://doi.org/10.1155/2012/215959.
29. Chang CH, Li WH, Shi YC, Huang CW, Chang ST, Liao VHC. *Caenorhabditis elegans* as model organisms to study the antioxidant potency of *Acacia confusa* twig extract. *Quart J Chin Forest*. 2012;45:503–514.
30. Adibhatla RM, Hatcher JF. Altered lipid metabolism in brain injury and disorders. *Subcell Biochem*. 2008;49:241–268.
31. Florence TM. The role of free radicals in disease. *Aust N Z J Ophthalmol*. 1995;23:3–7.
32. Tung YT, Wu JH, Huang CC, et al. Protective effect of *Acacia confusa* bark extract and its active compound gallic acid against carbon tetrachloride-induced chronic liver injury in rats. *Food Chem Toxicol*. 2009;47:1385–1392.

33. http://www.who.int/mediacentre/news/releases/2016/world-hepatitis-day/en/. Accessed 20 July 2016.
34. Thilakchand KR, Mathai RT, Simon P, Ravi RT, Baliga-Rao MP, Baliga MS. Hepatoprotective properties of the Indian gooseberry (*Emblica officinalis* Gaertn): a review. *Food Funct.* 2013;4:1431–1441.
35. Adewusi EA, Afolayan AJ. A review of natural products with hepatoprotective activity. *J Med Plants Res.* 2010;4:1318–1334.
36. Chattopadhyay RR. Possible mechanism of hepatoprotective activity of *Azadirachta indica* leaf extract: part II. *J Ethnopharmacol.* 2003;89:217–219.
37. Jain SK, Rajvaidy S, Desai P, Singh GK, Nagori BP. Herbal extract as hepatoprotective-a review. *J Pharmacogn Phytochem.* 2013;2:170–175.
38. Kumar A. A review on hepatoprotective herbal drugs. *Int J Res Pharm Chem.* 2012;2:96–102.
39. Kumar SV, Sanjeev T, Ajay S, Kumar SP, Anil S. A review on hepatoprotective activity of medicinal plants. *Int J Pharm Biosci.* 2012;2:31–38.
40. Bhawna S, Kumar SU. Hepatoprotective activity of some indigenous plants. *Int J Pharmtech Res.* 2009;4:1330–1334.
41. Tung YT, Chang ST. Inhibition of xanthine oxidase by *Acacia confusa* extracts and their phytochemicals. *J Agric Food Chem.* 2010;58:781–786.
42. Harris MD, Siegel LB, Alloway JA. Gout and hyperuricemia. *Am Fam Physician.* 1999;59:925–934.
43. Rasmussen JT, Rasmussen MS, Petersen TE. Cysteines involved in the interconversion between dehydrogenase and oxidase forms of bovine xanthine oxidoreductase. *J Dairy Sci.* 2000;83:499–506.
44. Wortmann RL. Management of hyperuricemia. In: McCarthy DJ, Koopman WJ, eds. *Arthritis and Allied Conditions.* twelfth ed. Philadelphia: Lea and Febiger; 1993:1807–1818.
45. Star VL, Hochberg M. Prevention and management of gout. *Drugs.* 1993;45:212–222.
46. Kelley WN. Antihyperuricemic drugs. In: Kelley WN, Harris ED, Ruddy S, Sledge CB, eds. *Textbook of Rheumatology*. Philadelphia: W. B. Saunders Co; 1991:862–877.
47. Lee TH, Liu DZ, Hsu FL, Wu WC, Hou WC. Structure-activity relationships of five myricetin galloylglycosides from leaves of *Acacia confusa*. *Bot Stud.* 2006;47:37–43.
48. Boomsma F, van Dijk J, Bhaggoe UM, Bouhuizen AMB, van den Meiracker AH. Variation in semicarbazide-sensitive amine oxidase activity in plasma and tissues of mammals. *Comp Biochem Physiol, C.* 2000;126:69–78.
49. Mullally MM, Meisel H, FitzGerald RJ. Synthetic peptides corresponding to α-lactalbumin and β-lactoglobulin sequences with angiotensin-I-converting enzyme inhibitory activity. *Biol Chem.* 1996;377:259–260.
50. Fotherby MD, Panayiotou B. Antihypertensive therapy in the prevention of stroke: what, when, and for whom? *Drugs.* 1999;58:663–674.
51. Wang SY, Yang CW, Liao JW, Zhen WW, Chu FH, Chang ST. Essential oil from leaves of *Cinnamomum osmophloeum* acts as a xanthine oxidase inhibitor and reduces the serum uric acid levels in oxonate-induced mice. *Phytomedicine.* 2008;15:940–945.
52. Zhu JX, Wang Y, Kong LD, Yang C, Zhang X. Effects of *Biota orientalis* extract and its flavonoid constituents, quercetin and rutin on serum uric acid levels in oxonate-induced mice and xanthine dehydrogenase and xanthine oxidase activities in mouse liver. *J Ethnopharmacol.* 2004;93:133–140.
53. de Heredia FP, Gomez-Martinez S, Marcos A. Obesity, inflammation and the immune system. *Proc Nutr Soc.* 2012;71:332–338.
54. Hotamisligil GS. Inflammation and metabolic disorders. *Nature.* 2006;444:860–867.
55. Sastre M, Klockgether T, Heneka MT. Contribution of inflammatory processes to Alzheimer's disease: molecular mechanisms. *Int J Dev Neurosci.* 2006;24:167–176.
56. Gorman C, Park A, Dell K. Health: the fires within. *Time.* 2004;163:38–45, 23 February, USA edition.
57. Lee JC, Chen WC, Wu SF, et al. Anti-hepatitis C virus activity of *Acacia confusa* extract via suppressing cyclooxygenase-2. *Antivir Res.* 2011;89:35–42.
58. Alter MJ. Epidemiology of hepatitis C virus infection. *World J Gastroenterol.* 2007;13:2436–2441.
59. Levrero M. Viral hepatitis and liver cancer: the case of hepatitis C. *Oncogene.* 2006;25:3834–3847.
60. Schaefer M, Mauss S. Hepatitis C treatment in patients with drug addiction: clinical management of interferon-alpha-associated psychiatric side effects. *Curr Drug Abuse Rev.* 2008;1:177–187.
61. Sun B, Karin M. NF-kappaB signaling, liver disease and hepatoprotective agents. *Oncogene.* 2008;27:6228–6244.
62. Lu L, Wei L, Peng G, et al. NS3 protein of hepatitis C virus regulates cyclooxygenase-2 expression through multiple signaling pathways. *Virology.* 2008;371:61–70.
63. Waris G, Siddiqui A. Hepatitis C virus stimulates the expression of cyclooxygenase-2 via oxidative stress: role of prostaglandin E2 in RNA replication. *J Virol.* 2005;79:9725–9734.
64. Núñez O, Fernández-Martinez A, Majano PL, et al. Increased intrahepatic cyclooxygenase 2, matrix metalloproteinase 2, and matrix metalloproteinase 9 expression is associated with progressive liver disease in chronic hepatitis C virus infection: role of viral core and NS5A proteins. *Gut.* 2004;53:1665–1672.
65. Penin F, Dubuisson J, Rey FA, Moradpour D, Pawlotsky JM. Structural biology of hepatitis C virus. *Hepatology.* 2004;39:5–19.
66. Flaherty DK. *Immunology for Pharmacy*. St. Louis: Mosby. Elsevier Inc; 2012.
67. LeBlanc DM, Barousse MM, Fidel Jr PL. Role for dendritic cells in immunoregulation during experimental vaginal candidiasis. *Infect Immun.* 2006;74:3213–3221.
68. Ho ST, Tung YT, Wu YJ, Lin CC, Wu JH. Immune-regulatory activity of methanolic extract of *Acacia confusa* heartwood and melanoxetin isolated from the extract. *Holzforschung.* 2015;69:645–652.
69. Dalod M, Chelbi R, Malissen B, Lawrence T. Dendritic cell maturation: functional specialization through signaling specificity and transcriptional programming. *EMBO J.* 2014;33:1104–1116.
70. Trucci VM, Salum FG, Figueiredo MA, Cherubini K. Interrelationship of dendritic cells, type 1 interferon system, regulatory T cells and toll-like receptors and their role in *Lichen planus* and *Lupus erythematosus* - a literature review. *Arch Oral Biol.* 2013;58:1532–1540.
71. Yamada H, Arai T, Endo N, et al. LPS-induced ROS generation and changes in glutathione level and their relation to the maturation of human monocyte-derived dendritic cells. *Life Sci.* 2006;78:926–933.
72. Sen P, Kemppainen E, Orešič M. Perspectives on systems modeling of human peripheral blood mononuclear cells. *Front. Mol. Biosci.* 2018. https://doi.org/10.3389/fmolb.2017.00096.
73. Kuo YC, Yang LM, Lin LC. Isolation and immunomodulatory effect of flavonoids from *Syzygium samarangense*. *Planta Med.* 2004;70:1237–1239.
74. Kang JH, Lee J, Moon M, Yim M. 3'4'7-Trihydroxyflavone inhibits RANKL-induced osteoclast formation via NFATc1. *Pharmazie.* 2015;70:661–667.
75. Kang JH, Jung H, Yim M. 3',4',7,8-Tetrahydroxyflavone inhibits RANKL-induced osteoclast formation and bone resorption. *Pharmazie.* 2017;72:161–166.
76. Yavropoulou MP, Yovos JG. Osteoclastogenesis - current knowledge and future perspectives. *J Musculoskelet Neuronal Interact.* 2008;8:204–216.
77. Takayanagi H, Kim S, Koga T, et al. Induction and activation of the transcription factor NFATc1 (NFAT2) integrate RANKL signaling in terminal differentiation of osteoclasts. *Dev Cell.* 2002;3:889–901.
78. Novack DV, Teitelbaum SL. The osteoclast: friend or foe? *Ann Rev Pathol.* 2008;3:457–484.
79. Shulgin A, Shulgin A. *TIHKAL: the Continuation*. Berkeley: Transform Press; 1997.
80. Strassman RJ, Qualls CR. Dose-response study of *N,N*-dimethyltryptamine humans: I. neuroendocrine autonomic, and cardiovascular effects. *Arch Gen Psychiatr.* 1994;51:85–97.
81. Strassman RJ, Qualls CR, Uhlenhuth EH, Kellner R. Dose-response study of *N,N*-dimethyltryptamine in humans: II. subjective effects and preliminary results of a new rating scale. *Arch Gen Psychiatr.* 1994;51:98–108.
82. Stoff DM, Moja EA, Gillin JC, Wyatt RJ. Dose response and time course effects of *N,N*-dimethyltryptamine on disruption of rat shuttle box avoidance. *Biol Psychiatr.* 1977;12:339–346.
83. Carbonaro TM, Gatch MB. Neuropharmacology of *N,N*-dimethyltryptamine. *Brain Res Bull.* 2016;126:74–88.
84. Lowe LM, Peterson BL, Couper FJ. A case review of the first analytically confirmed 25I-NBOMe-related death in Washington State. *J Anal Toxicol.* 2015;39:668–671.
85. Hill SL, Doris T, Gurung S, et al. Severe clinical toxicity associated with analytically confirmed recreational use of 25I-NBOMe: case series. *Clin Toxicol.* 2013;51:487–492.
86. Strassman RJ. *DMT: the Spirit Molecule. A Doctor's Revolutionary Research into the Biology of Near-death and Mystical Experiences*. Rochester: Park Street Press; 2001.