Phytopharmacological Investigations of *Piper retrofractum* Vahl. – A Review

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Summary

Piper retrofractum Vahl. which belongs to the family Piperaceae, is a popular spice possessing high medicinal properties. The present article aims to provide a review of the studies done on phytochemistry and biological activities of *P. retrofractum*. This review is based on a literature study of scientific journals from electronic sources, such as Science Direct, PubMed, Google Scholar, Scopus, and Web of Science. The main chemical constituents that have been isolated and identified from *P. retrofractum* are amides, alkaloids, phenylpropanoids, alkyl glycosides, and lignans. This plant possesses antioxidant, hepatoprotective, cytotoxic, larvicidal, antiproliferation, antitubercular, antileishmanial, antiphotoaging, and anti-obesity properties. *P. retrofractum* has the potential for the treatment of several diseases and disorders, but there are only a few studies done to investigate the plant phytochemicals, thus further studies should be focused on isolation and identification of active compounds with pharmacological activities. Besides, the majority of pharmacological studies have been performed using aerial parts of the plant, however, further studies are needed to investigate bioactivity of other parts of the plant.

Key words

Piperaceae, *Piper*, phytochemicals, pharmacology, amide, alkaloid

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Introduction

Tropical regions have been known as the home of many fascinating medicinal plant species. This includes medicinal plants from the genus *Piper* that covers around one thousand species. In terms of morphological characteristics, all species in this genus possess three plant forms including creeping, climbing, and branching stems. In addition, all plants have distinct leaves color and shape. Apart from its role in pharmaceutical domains, plants from the genus *Piper* are also used by traditional communities as supporting material for decorative arts, traditional ceremonies as well as food, and beverages (Chaveerach et al., 2006). The chemistry of *Piper* species has been widely investigated and the phytochemical investigations from all parts have led to the isolation of a number of pharmacologically active compounds. They have been extensively investigated as a source of new natural products with potential antioxidant, antimicrobial, antifungal, anticholinesterase, anti-inflammatory, anti-tyrosinase, and insecticidal activities (Salleh et al., 2012, 2014a, 2014b, 2014c, 2014d, 2015a, 2015b, 2016a, 2016b, 2019).

Piper retrofractum Vahl. (syn. *Piper chaba* Hunter) is a member of the Piperaceae family and traditionally known as *Javanese long pepper.* The stem can reach up to 12 m length. The leaves are short-petioled, green in color, pale when dry, oblong-ovate or elliptic-lanceolate in shape, 6 to 7.5 cm long and 3 to 8 cm wide. The base is sublateral or inequilateral, pointed or slightly cordate with tips acute. The leaves are penninerved, 7 to 11 lateral nerves on each side of the midrib, ascending. The male spikes are 3.8 to 8.5 cm long and 2.5 to 4.5 mm in diameter; the bracts somewhat stalked and peltate. There are 2 to 3 stamens, which are stalkless. The female spikes are oblong when matured, red in color, fleshy, cylindrical, 3 to 6.5 cm long and 6.5 to 11 mm in diameter, the rachis is smooth and the bracts are stalkless and peltate. The fruit is more or less united partly or wholly embedded in and concrescent with rachis. There are three short stigmas. The seeds are subglobose to obovoid-globose, 2 to 2.5 mm long (Chopra et al., 1986). The plant is an outstanding pharmaceutical species that is native to Southeast Asia and predominantly cultivated in Indonesia, Thailand, Malaysia, Bangladesh, Vietnam, and India. It has been used as a source of medicinal compounds to treat various conditions. Besides, the plant is commonly used as a spice in cooking (Lim, 2012). Several studies have been performed to reveal the pharmacological potentials of *P. retrofractum* during the last decade.

This review aims to summarize the available information on their traditional uses, phytochemicals, and biological activities. The literature used in the review comprises scientific journals from electronic sources, such as Science Direct, PubMed, Google Scholar, Scopus, and Web of Science. Chemical structures presented in this article are either drawn with ACD/ChemSketch or obtained from the literature.

Traditional Uses

Available literature and information show that *P. retrofractum* has been applied as traditional medicines in various parts of Asia. The roots of *P. retrofractum* can act as stimulants and are used to treat asthma, bronchitis, hemorrhoids, fever, liver diseases, jaundice, edema, and abdominal pain. Both the unripe and ripe fruits of *P. retrofractum* have been used as a spice in curries, preservatives, and pickles (Lim, 2012). In Thailand, this plant has been categorized nationally as an essential drug for the treatment of the gastrointestinal system, while the fruit is useful for treating bronchitis, bronchial asthma, and muscle pain (Farnsworth and Bunyapraphatsara, 1992). Traditionally, the fruits of *P. retrofractum* are used in Indonesian folk medicine (*Jamu*) as a tonic against a variety of digestive, stimulant, carminative, intestinal disorders and for treating postpartum women (Lim, 2012). In Bangladesh, the leaves are used to treat carotid artery disease and tendon discomfort. The root is applied to treat paresis, yaws, and diarrhea, and is used as an antipyretic and carminative (Yusuf et al., 1994). Meanwhile, in Japan, the dried fruits are traditionally used as a seasoning, which has a unique pungent taste and aroma (Takahashi et al., 2017). In Ayurveda, the plant is traditionally used to promote respiratory and digestive health, and it is an ingredient in medicinal teas (Lim, 2012). Based on the traditional use, *P. retrofractum* is a potential component for herbal medicine development, therefore, its pharmaceutical analysis is necessarily needed.

Phytochemicals

A review of the literature revealed that few phytochemical studies have been carried out on *P. retrofractum* prior to the current study. Up to now, 97 compounds (besides volatile constituents) have been reported including seventy-seven amides (Banerji et al., 1985; Ahn et al., 1992; Kikuzaki et al., 1993; Bodiwala et al., 2007; Kubo et al., 2013; Muharini et al., 2015; Amad et al, 2017; Tang et al., 2019), four amide glycosides (Tang et al., 2019), eleven alkyl glycosides, one phenylpropanoid (Luyen et al., 2014), two phenylpropanoid glycosides (Hieu et al., 2014), and two lignans (Banerji et al., 1985). Among them, twenty-seven new phytochemicals have been isolated from various parts of *P. retrofractum* and their chemical structures are shown in Figure 1. The list of phytochemicals of *P. retrofractum* is shown in Table 1.

Amides are the predominant secondary metabolite constituents in this species and mostly unsaturated amides. A total of 18 new amides have been successfully identified. Amide glycosides have also been reported that are retrofractosides A-D (Tang et al., 2019). To date, eleven alkyl glycosides were isolated from the leaves of *P. retrofractum* as reported by Luyen et al. (2014). They have successfully isolated a new compound, isoheptanol-2(S)-O-β-D-xylopyranosyl (1→6)-O-β-D-glucopyranoside. In addition, a new phenylpropanoid, piperoside was also obtained from the leaves of *P. retrofractum*. Meanwhile, two phenylpropanoid glycosides have been reported from the fruits of this species. Retrofractosides E-F (Tang et al., 2019). Furthermore, the only lignans isolated from this species were sesamin and 3,4,5-trimethoxydihydrocinnamic acid (Banerji et al., 1985). Additionally, two studies have been performed investigating the essential oil composition of *P. retrofractum*. Hieu et al. (2014) reported the leaf oil collected from Vietnam. The main classes of compounds were monoterpene hydrocarbons (34.4%) and benzyl benzoate (14.4%) as the major component. In addition, Hao et al. (2018) were successfully identified 21 components from the leaf oil, collected from China. The oil shows its richness in ocimene (15.6%) and linalool (12.9%).

Jadid et al. (2018) investigated the proximate and mineral composition of the fruits of *P. retrofractum*. The fruits contained carbohydrate (63.4%), crude protein (11.4%), total ash (4.29%), dietary fiber (28.8%) and total fat (2.97%).

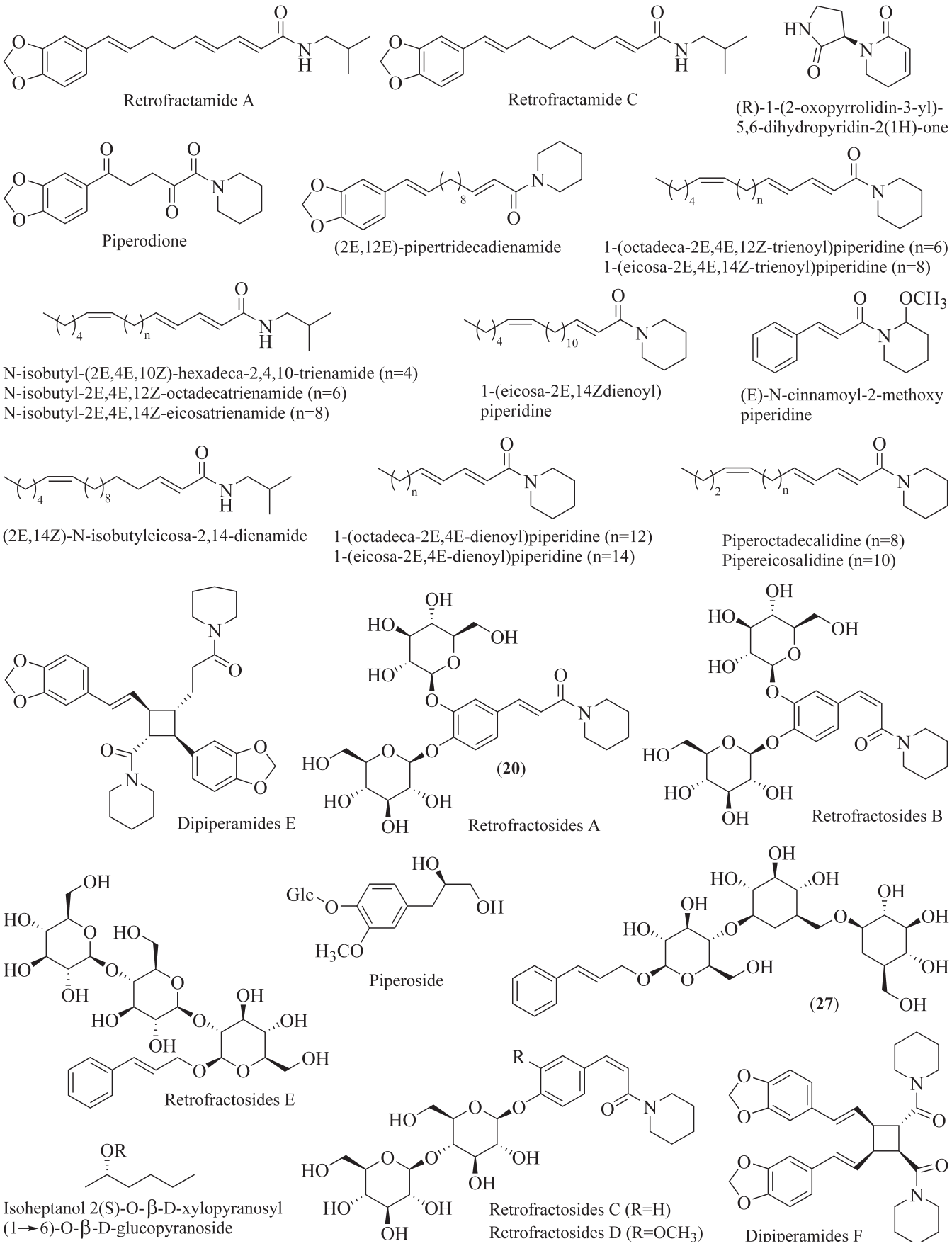

Figure 1. Chemical structures of several new phytochemicals isolated from *Piper retrofractum*

Table 1. Phytochemicals isolated from *Piper retrofractum*

Phytochemicals	Plant parts	References
AMIDES		
Retrofractamide A	Aerial	Banerji et al., 1985
	Fruits	Muharini et al., 2015
	Fruits	Kubo et al., 2013
Retrofractamide B	Aerial	Banerji et al., 1985
	Fruits	Kubo et al., 2013
	Fruits	Muharini et al., 2015
	Fruits	Tang et al., 2019
	Fruits	Kikuzaki et al., 1993
Retrofractamide C	Aerial	Banerji et al., 1985
	Fruits	Muharini et al., 2015
	Fruits	Kubo et al., 2013
Retrofractamide D	Roots	Banerji et al., 2002
Piperine	Fruits	Kubo et al., 2013
	Fruits	Amad et al., 2017
	Fruits	Muharini et al., 2015
	Fruits	Ahn et al., 1992
	Fruits	Kikuzaki et al., 1993
	Fruits	Tang et al., 2019
	Fruits	Bao et al., 2014
Methyl piperate	Fruits	Amad et al., 2017
	Fruits	Kikuzaki et al., 1993
	Fruits	Tang et al., 2019
	Fruits	Musthapa et al., 2018
Piperlonguminine	Fruits	Kubo et al., 2013
	Fruits	Amad et al., 2017
	Fruits	Tang et al., 2019
	Fruits	Kikuzaki et al., 1993
	Fruits	Bao et al., 2014
Dihydropiperlonguminine	Fruits	Muharini et al., 2015
	Fruits	Tang et al., 2019
Guineensine	Fruits	Kikuzaki et al., 1993
	Fruits	Muharini et al., 2015
	Fruits	Tang et al., 2019
Sylvatine	Fruits	Amad et al., 2017

Phytochemicals	Plant parts	References
Pellitorine	Stems	Bodiwala et al., 2007
	Fruits	Muharini et al., 2015
	Fruits	Kikuzaki et al., 1993
Piplartine	Stems	Bodiwala et al., 2007
Piperodione	Fruits	Kubo et al., 2013
(*2E,12E*)-pipertridecadienamide	Fruits	Kubo et al., 2013
N-isobutyl-(*2E,4E,10Z*)-hexadeca-2,4,10-trienamide	Fruits	Kubo et al., 2013
Isopiperine	Fruits	Kubo et al., 2013
Isochavicine	Fruits	Kubo et al., 2013
5,6-Dihydropiperlongmine	Fruits	Kubo et al., 2013
5,6-Dihydro-1*H*-pyridin-2-one	Fruits	Tang et al., 2019
cis-Fragaramide	Fruits	Kubo et al., 2013
trans-Fragaramine	Fruits	Kubo et al., 2013
	Fruits	Tang et al., 2019
N-isobutyl-(2E,4E,12Z)-octadeca-2,4,12-trienamide	Fruits	Tang et al., 2019
N-isobutyl-(2E,4E,12Z)-hexadeca-2,4,12-trienamide	Fruits	Kubo et al., 2013
N-isobutyl-(2E,4E)-hexadeca-2,4-dienamide	Fruits	Kubo et al., 2013
(*2E,4Z,8E*)-*N*-[(3,4-methylendioxy-phenyl)-2,4,8-nonatrienoyl]piperidine	Fruits	Kubo et al., 2013
N-Cinnamoylpiperidine	Fruits	Kubo et al., 2013
Piperoleine B	Fruits	Kubo et al., 2013
	Fruits	Tang et al., 2019
Piperchabamide B	Fruits	Kubo et al., 2013
	Fruits	Tang et al., 2019
Piperchabamide C	Fruits	Kubo et al., 2013
	Fruits	Muharini et al., 2015
Piperchabamide H	Fruits	Kubo et al., 2013
	Fruits	Muharini et al., 2015
Piperundecaline	Fruits	Kubo et al., 2013
	Fruits	Muharini et al., 2015
Dehydropipernonaline	Fruits	Kubo et al., 2013
	Fruits	Muharini et al., 2015
Pipernonaline	Fruits	Kubo et al., 2013
	Fruits	Muharini et al., 2015
	Fruits	Ahn et al., 1992
	Fruits	Tang et al., 2019
N-isobutyl-*2E,4E,12Z*-octadecatrienamide	Fruits	Kikuzaki et al., 1993

Phytochemicals	Plant parts	References
N-isobutyl-*2E,4E,14Z*-eicosatrienamide	Fruits	Kikuzaki et al., 1993
1-(octadeca-*2E,4E,12Z*-trienoyl)piperidine	Fruits	Kikuzaki et al., 1993
1-(eicosa-*2E,4E,14Z*-trienoyl)piperidine	Fruits	Kikuzaki et al., 1993
1-(octadeca-*2E,4E*-dienoyl)piperidine	Fruits	Kikuzaki et al., 1993
1-(eicosa-*2E,4E*-dienoyl)piperidine	Fruits	Kikuzaki et al., 1993
1-(eicosa-*2E,14Z*dienoyl)piperidine	Fruits	Kikuzaki et al., 1993
(*E*)-*N*-cinnamoyl-2-methoxypiperidine	Fruits	Tang et al., 2019
(*R*)-1-(2-oxopyrrolidin-3-yl)-5,6-dihydropyridin-2(1*H*)-one	Fruits	Tang et al., 2019
(*E*)-*N*-(Tetrahydro-*2H*-pyran-2-yl)cinnamamide	Fruits	Tang et al., 2019
Piperanine	Fruits	Tang et al., 2019
3-Phenyl-1-(piperidin-1-yl)propan-1-one	Fruits	Tang et al., 2019
(*E*)-3-Phenyl-1-(piperidin-1-yl)prop-2-en-1-one	Fruits	Tang et al., 2019
3-Chloro-4-hydroxy-2-piperidone	Fruits	Tang et al., 2019
Octahydro-4-hydroxy-3α-methyl-7-methylene-α-(1-methylethyl)-1H-indene-1-methanol	Fruits	Tang et al., 2019
Alismoxide	Fruits	Tang et al., 2019
(4*S*,4a*S*,6*S*,8a*S*)-octahydro-4-hydroxy-4,8a-dimethyl-6-(1-methylethenyl)-naphthalen-1(*2H*)-one	Fruits	Tang et al., 2019
ent-4(15)-Eudesmene-1β,6α-diol	Fruits	Tang et al., 2019
Methylsalicylate-2-*O*-β-D-glucopyrano-side	Fruits	Tang et al., 2019
Rozin	Fruits	Tang et al., 2019
Piperchabaoside A	Fruits	Tang et al., 2019
(6*S*,9*R*)-Roseoside	Fruits	Tang et al., 2019
2-*O*-Methyluridine	Fruits	Tang et al., 2019
(2*E*,14*Z*)-*N*-isobutyleicosa-2,14-dienamide	Fruits	Muharini et al., 2015
Dipiperamides E	Fruits	Muharini et al., 2015
Dipiperamides F	Fruits	Muharini et al., 2015
Dipiperamides G	Fruits	Muharini et al., 2015
(*E*)-4-(isobutylamino)-4-oxo-2-butenoic acid	Fruits	Muharini et al., 2015
3,4-Methylenedioxycinnamaldehyde	Fruits	Muharini et al., 2015
Piperonyl anhydride	Fruits	Muharini et al., 2015
Isochavicine	Fruits	Muharini et al., 2015
Scutifoliamide A	Fruits	Muharini et al., 2015
Pipericine	Fruits	Muharini et al., 2015
(2*E*,4*E*)-*N*-isobutyleicosa-2,4-dienamide	Fruits	Muharini et al., 2015
(2*E*,4*E*,12*Z*)-*N*-isobutyloctadec-2,4,12-trienamide	Fruits	Muharini et al., 2015
(2*E*,4*E*,14*Z*)-*N*-isobutyleicosa-2,4,14-eicosatrienamide	Fruits	Muharini et al., 2015

Phytochemicals	Plant parts	References
Pipereicosalidine	Fruits	Muharini et al., 2015
Brachystamide B	Fruits	Muharini et al., 2015
Piperolein B	Fruits	Muharini et al., 2015
Chabamide	Fruits	Muharini et al., 2015
Nigramide F	Fruits	Muharini et al., 2015
Nigramide R	Fruits	Muharini et al., 2015
Piperoctadecalidine	Fruits	Ahn et al., 1992
Pipereicosalidine	Fruits	Ahn et al., 1992
3-Methyl-5-decanoylpyridine	Fruits	Pande et al., 1997
AMIDE GLUCOSIDES		
Retrofractosides A	Fruits	Tang et al., 2019
Retrofractosides B	Fruits	Tang et al., 2019
Retrofractosides C	Fruits	Tang et al., 2019
Retrofractosides D	Fruits	Tang et al., 2019
ALKYL GLYCOSIDES		
3,4-Dihydroxyallylbenzene	Leaves	Luyen et al., 2014
1,2-di-*O*-*β*-D-Glucopyranosyl-4-allylbenzene	Leaves	Luyen et al., 2014
Tachioside	Leaves	Luyen et al., 2014
Benzyl-O-β-D-glucopyranoside	Leaves	Luyen et al., 2014
Icariside F_2	Leaves	Luyen et al., 2014
Dihydrovomifoliol-*O*-*β*-D-glucopyranoside	Leaves	Luyen et al., 2014
Isopropyl *O*-*β*-D-glucopyranoside	Leaves	Luyen et al., 2014
Isopropyl primeveroside	Leaves	Luyen et al., 2014
n-Butyl *O*-*β*-D-glucopyranoside	Leaves	Luyen et al., 2014
Isoheptanol 2(S)-*O*-*β*-D-xylopyranosyl (1→6)-*O*-*β*-D-glucopyranoside	Leaves	Luyen et al., 2014
Isoheptanol 2(S)-*O*-*β*-D-apiofuranosyl-(1→6)-*O*-*β*-D-glucopyranoside	Leaves	Luyen et al., 2014
PHENYLPROPANOID GLYCOSIDES		
Retrofractosides E	Fruits	Tang et al., 2019
Retrofractosides F	Fruits	Tang et al., 2019
PHENYLPROPANOID		
Piperoside	Leaves	Luyen et al., 2014
LIGNANS		
Sesamin	Aerial	Banerji et al., 1985
3,4,5-Trimethoxydihydrocinnamic acid	Aerial	Banerji et al., 1985

The fruit also contained calcium, copper, iron, magnesium, phosphor, potassium, sodium, and zinc in different concentrations. Additionally, quinone, sterol, glycosides, and alkaloids were detected in both *n*-hexane and ethyl acetate extracts. Tannin was present in ethyl acetate and methanol extracts. Methanol extract contained sterol, glycosides, flavones, tannin, and alkaloids. The results also revealed that methanol extract of the fruit contained the highest phenol compared to other extracts.

Pharmacology

The literature study revealed the need for a thorough investigation of the pharmacological characteristics of the extracts and isolated compounds from *P. retrofractum*. The biological activities, including antioxidant (Wasito et al., 2011; Jadid et al., 2017; Mahaldar et al., 2019), hepatoprotective (Mahaldar et al., 2019), cytotoxic (Amad et al., 2017; Ekowati et al., 2012), larvicidal (Chansang et al., 2005), antiproliferation (Hasan et al., 2016), antitubercular (Amad et al., 2017), antileishmanial (Bodiwala et al., 2007), antiphotoaging (Yun et al., 2018), and anti-obesity (Kim et al., 2011) activities, have been reported in several research articles. Indeed, *P. retrofractum* has been exploited traditionally, thus revealing the medicinal variation it possesses. This information on the qualification of the extracts is essential in view of their potential applications in functional food and pharmaceutical areas.

Antioxidant activity

Jadid et al. (2017) reported the antioxidant activity of *P. retrofractum* fruit extracted in different solvents (methanol, ethyl acetate, *n*-hexane) using 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay at various concentrations. The results showed that the DPPH free radicals were scavenged by all plant extracts in a concentration-dependent manner. The IC_{50} values for DPPH radicals with methanol, ethyl acetate, and *n*-hexane extract of the *P. retrofractum* were found to be 101.74, 66.12, and 57.66 ppm, respectively. Interestingly, the IC_{50} value of *n*-hexane extract (57.66 ppm) was lower than ascorbic acid (66.12 ppm), indicating that *n*-hexane extract was a more potent scavenger of free radicals than methanol and ethyl acetate extracts. The root and stem ethanol extracts were also investigated by *in vitro* antioxidant activity (Mahaldar et al., 2019). In DPPH free radical assay the ethanol extract of *P. retrofractum* stem and root showed IC_{50} values 133 mg/mL and 91 mg/mL, respectively, while for ascorbic acid it was 14 mg/mL. In hydroxyl radical scavenging assay the stem and root extracts exhibited IC_{50} at 189 mg/mL and 144 mg/mL, respectively. The root extract demonstrated the highest reducing power (RC_{50}: 172.59 mg/mL) in comparison to the stem extract (RC_{50}: 274.85 mg/mL). Reducing power of both extracts exhibited concentration-dependency and was comparable with standard ascorbic acid (RC_{50}: 120.95 mg/mL). Wasito et al. (2011) reported that antioxidant activity of the ethanolic extract of *P. retrofractum* produced an IC_{50} of 3.445 µg/mL. The IC_{50} of a combination of Zingiber officinale and *P. retrofratum* ethanolic extracts at concentration ratios of 1:2, 1:1, and 2:1 were 148 µg/mL, 85 µg/mL, and 73 µg/mL, respectively.

Hepatoprotective activity

Hepatoprotective activity was reported against paracetamol-induced acute hepatotoxicity and estimated in Sprague-Dawley rats. The activity was evaluated by measuring serum biomarker enzymes (SGPT and SGOT), serum biochemical parameters (total proteins, albumin, and bilirubin), and serum lipid profiling (cholesterol, HDL-c, and LDL-c). Ethanol extracts of *P. retrofractum* showed significant hepatoprotection against paracetamol-induced hepatotoxicity in rats by reducing serum total bilirubin ($p < 0.001$), GPT ($p < 0.01$), and GOT ($p < 0.01$). Both stem and root extracts exerted a beneficial effect on total protein, albumin, and serum lipid profile. Meanwhile, in the histopathological study of liver sections, both extracts showed minimal to mild multifocal and diffuse granular degeneration and mild to moderate lobular disarray compared to the control group. The authors suggest that both extracts can prevent paracetamol-induced hepatotoxicity (Mahaldar et al., 2019).

Cytotoxic activity

The crude methanol extract showed moderate cytotoxic behavior against lung cancer cells (NCI-H187) and human gingival fibroblasts (HGF) (Amad et al., 2017). Furthermore, the cytotoxic and apoptotic activity against p53 expression on Myeloma and WiDr cells has been reported (Ekowati et al., 2012). Results showed that *P. retrofractum* extract and its combination with *Zingiber officinale* had cytotoxic activity on Myeloma cells with IC_{50} of 36 and 55 mg/mL, respectively, and WiDr cell lines with IC_{50} of 158 and 64 mg/mL, respectively. Extracts also induced apoptotic activity, and increased p53 expression on Myeloma and WiDr cells.

Larvicidal activity

Chansang et al. (2005) reported the mosquito larvicidal activity of the fresh fruits water extract. Extracts of unripe and ripe fruits showed different levels of activity against *Culex quinquefasciatus* and *Aedes aegypti* larvae. The ripe fruit extract was more active against *Aedes aegypti* than *Culex quinquefasciatus*. Another ripe fruit extract was much more toxic to both mosquito species. Wiwattanawanichakun et al. (2018) investigated the acute toxicity of crude extracts and alkaloid compounds of *P. retrofractum* in *Culex quinquefasciatus* third instar larvae. The results showed crude hexane extract had the highest toxicity for *Culex quinquefasciatus* (0.9 ppm), whereas piperine and piperanine showed LC_{50} values of 0.27 and 2.97 ppm, respectively, after 24 h of exposure.

Antiproliferation activity

The anti-proliferation activity of the *P. retrofractum* fruits extract against breast cancer cells MCF-7 has been reported (Hasan et al., 2016). Five samples of the *P. retrofractum* fruits were collected from Central Java and Lampung, in Indonesia. The results revealed that the extracts from Lampung displayed significant inhibition of cells with IC_{50} value of 4.35 µg/mL, compared to doxorubicin with IC_{50} value of 58.7 µg/mL.

Antitubercular activity

The crude methanol extract showed antibacterial activity against *Mycobacterium tuberculosis*, with a minimum inhibitory concentration (MIC) of 25.00 µg/mL. The activity was assessed by measuring the green fluorescent protein (GFP) produced by the GFP expressing *M. tuberculosis* strain H37Ra. Isoniazid, rifampicin, ethambutol, ofloxacin, and streptomycin were used as positive controls and gave MIC of 0.023-0.047, 0.003-0.025, 0.234-0.469, 0.391-0.781, and 0.156-0.313, respectively (Amad et al., 2017).

Antileishmanial activity

The *n*-hexane, ethyl acetate, methanol, and acetone extracts of *P. retrofractum* were evaluated *in vitro* against promastigotes of *Leishmania donovani*, and all exhibited significant *in vitro* activity. The methanol and acetone extracts showed more than 75% inhibition at a concentration of 20 µg/mL with IC_{50} values of 7.5 and 3.5 µg/mL, respectively. However, (-)-sesamin was found inactive (Bodiwala et al., 2007).

Antiphotoaging effects

P. retrofractum extract has shown antiphotoaging activities in UVB-damaged human dermal fibroblasts and skin of hairless mice via PPARδ and AMPK agonist activities. As a PPARδ and AMPK activator, the extract induced mitochondrial biogenesis that was suppressed by UVB-irradiation. Additionally, the extract suppressed MMPs expression through PPARδ/AMPK activation and MAPKs/AP-1 inhibition, while enhancing collagen gene expression by upregulating TGF-δ and Smad signaling mediated by PPARδ activation. The extract alleviated inflammatory responses by suppressing NF-κB activity. *P. retrofractum* extract prevented histological deformations, such as wrinkle formation, ECM destruction, and erythema mediated by UVB-irradiation in the skin of hairless mice (Yun et al., 2018).

Anti-obesity activity

Kim et al. (2011) evaluated the anti-obesity effects of *P. retrofractum* in high-fat diet (HFD)-induced obese mice. They found that in the animal model, oral extract administration (50, 100, or 300 mg/kg/day for eight weeks) significantly reduced HFD-induced body weight gain without altering the amount of food intake. In addition, elevated serum levels of total cholesterol, low-density lipoprotein cholesterol, total lipid, leptin, and lipase were suppressed by extract treatment. The extract also protected against the development of non-alcoholic fatty liver by decreasing hepatic triglyceride accumulation. Consistent with the *in vitro* results, the extract activated AMPK signaling and altered the expression of lipid metabolism-related proteins in liver and skeletal muscle.

Conclusion

Several studies have been carried out focusing on the pharmacological activities of *P. retrofractum*. These studies make it possible to justify the traditional uses of the plant and to find new pharmacological activities. However, the chemical composition of the plant has not been extensively studied and deserves more attention in the near future because the phytochemicals analysis of the plants is a key component of medicinal plant research and drug discovery. Therefore, there is a need for more phytochemical studies using different parts of the plants and isolation of active compounds that should be tested for their pharmacological activities.

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