Chapter

Calophyllum inophyllum: Beneficial Phytochemicals, Their Uses, and Identification

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Abstract

Calophyllum inophyllum Linn. is one type of mangrove plant. This plant is commonly called nyamplung. This plant is abundant in Indonesia and has many properties that can be exploited from the roots, stems, and leaves to the seeds. All parts of this plant can be useful for human needs. Its oil is generally only used as biodiesel feedstock. The aim of this chapter is to discuss the identification and the uses of phytochemicals contained in *C. inophyllum* leaves. There are various kinds of phytochemicals contained in *C. inophyllum* leaves, such as triterpenoids, steroids, flavonoids, coumarins, xanthones, fatty acids, esters, alkenes, ethers, and alicyclic compounds. They have benefits to health, such as anticancer, anti-HIV, antiviral, antitumor, anti-inflammatory, antimicrobial, antineoplastic, antiplatelet, antipsychotics, antioxidant, antiaging, antileukemic, antimalarial, anticoagulant, antifeedant, analgesic, photoprotective, molluscicidal, and piscicidal agents. Extraction is a famous method for isolating phytochemicals in *C. inophyllum* leaves, based on the solvent polarity index.

Keywords: *C. inophyllum*, human health, identification, isolation, phytochemicals

1. Introduction

The name of Calophyllum inophyllum is Kallos that is taken from the Greek word, which means beautiful and meaningful Phullon leaves. *C. inophyllum* has many name designations that vary by region country. In the UK, the tree is known as a beautiful leaf (translation from Greek), Indian laurel (because it comes from India), Alexandrian laurel, and beach *Calophyllum* (because the trees usually grow on the waterfront). Moreover, the tree is also called as tamanu (Tahiti), fetau (Samoa), damanu (Fiji Island), te itai (Kiribati Island), nyamplung (Indonesia), Penaga Laut (Malaysia), kamani (Hawaii), foraha (Madagascar), and puna (island of Lakshadweep) [1].

According to Ong [2], the distribution map of *C. inophyllum* in the world is quite extensive. This species is commonly found in areas with a tropical climate. In the world, this species is found in countries such as Australia, Cambodia, the Cook Islands, Fiji, French Polynesia, India, Indonesia, Japan, Kiribati, Laos, Madagascar, Malaysia, the Marshall Islands, Myanmar, New Caledonia, Norfolk Island, Papua New Guinea, the Philippines, Reunion, Samoa, Solomon Islands, Sri Lanka, Taiwan, Province of China, Thailand, Tonga, Vanuatu, and Vietnam.

As for exotic species (endemic to a region), it can be found in the state of Djibouti, Eritrea, Ethiopia, Kenya, Nigeria, Somalia, Tanzania, Uganda, and the USA.

C. inophyllum plant spreads almost evenly throughout Indonesia, such as in the island of Sumatra (West Sumatra, Riau, Kepulauan Riau, Lampung, and Bangka Belitung), Java (Banten, West Java, Central Java, Yogyakarta, East Java), Bali Island, East Nusa Tenggara and West Nusa Tenggara, Kalimantan (West Kalimantan, Central Kalimantan, and South Kalimantan), Sulawesi (North Sulawesi, Gorontalo, Central Sulawesi, South Sulawesi, and Southeast Sulawesi), Maluku and North Maluku Islands, and Papua [3]. *C. inophyllum* plant has a taxonomy as follows [4]:

Kingdom: Plantae
Subkingdom: Tracheobionta
Super division: Spermatophyta
Division: Magnoliophyta
Class: Magnoliopsida
Subclass: *Dilleniidae*
Order: *Theales*
Family: Clusiaceae

C. inophyllum is a plant that is grown in the earthy sand and coastal areas with a hot weather [5]. It can also grow well at an altitude of 0–800 meters above sea level such as in forests, mountains, and swamps [6]. *C. inophyllum* is a versatile crop; all parts of this plant, such as leaves, root, and fruit (**Figure 1**), can be useful for humans. The benefit of its tree, bark, and seed is as plant conservation, source of timber and non-timber forest products (NTFPs), and vegetable oil, respectively [7]. In pharmaceuticals, it is known to function as an antibacterial, anticancer, antineoplastic, anti-inflammatory, antiplatelet, antipsychotics, antiviral, photoprotective, molluscicidal, and piscicidal agent [1]. **Table 1** shows the benefits of *C. inophyllum* crops obtained from previous works.

Because all parts of this plant can be useful in treating various diseases, some researchers have conducted further research on the phytochemical content of this plant. According to Ling et al. [1], the compounds which are contained in these plants include inophynone; canophyllol; canophyllic acid; calophyllolide;

Figure 1.
Parts of C. inophyllum crop.

Part of crops	Medicinal function		
	Iskandari and Anna [8]	**Su et al.** [9]	**Ling et al.** [1]
Leaves	Inhibit the growth of larvae of *Culex quinquefasciatus* and *Aedes aegypti*, an inhibitor of the HIV virus	Treat skin rashes, swelling of the legs, caring for burns, eye irritation, dysentery, migraine, and vertigo	Treat skin diseases, arthritis, sciatica, eye irritation
Root	Antibacterial	Treat dysentery, gonorrhea, indigestion, wounds, ulcers, and others	Treating internal hemorrhage
Fruit/ seed	Inhibit the growth of larvae of *Culex quinquefasciatus*, antimicrobial compounds, and toxic agents	Treating stomach pain, itching, arthritis, burns, gonorrhea, arthritis, ulcers, and ringworm	Treat wounds, leprosy, neurological diseases, burns

Table 1.
Benefits and uses of C. inophyllum crops.

inophyllolide; inophyllum B, C, P, and E; jacareubin; (+)-calanolide A; inocalophyllins A and B; calophynone; calophyllumin C; inophyllin A; and others. Su et al. [9] mentioned that according to Filho et al. [10], in various parts of the tree, C. inophyllum contains phytochemicals, including xanthones, coumarins, chromanones (flavonoids, biflavonoids), triterpenes, tripenoids, and steroids. Coumarins in *C. inophyllum* contain two components, namely, calanolides A and B. From these studies it was found that coumarin compounds in *C. inophyllum* may be effective in treating cancer and inhibiting the HIV virus.

According to Lim [11], at least nine components have been isolated from the leaves of *C. inophyllum*, including 2-hydroxyxanthone; 4-hydroxyxanthone; 1,5-dihydroxyxanthone; 1,7-dihydroxyxanthone; 1,3,5-trihydroxy-2-methoxyxanthone; 6-6-deoxyjacaerubin; flavonoids, amentoflavone; kaempferol-3-O-α-L-rhamnoside; and quercetin-3-O-α-L-rhamnoside.

Of the three studies on the leaves above, there are some differences as well as questions obtained from the leaves of *C. inophyllum* content analysis. Some of the same compounds that have been isolated from *C. inophyllum* plants are quite diverse, including derivatives of xanthones [12, 13], coumarins [9], flavonoids [13], benzodipyranones [14], triterpenoids [12, 15], and steroids [9].

2. Identification of phytochemicals in *C. inophyllum* leaves

2.1 Xanthones

Xanthones are polyphenol components in nature with molecular formula $C_{13}H_8O_2$. They consist of bonding of two benzene rings connected by a carbonyl group and one oxygen. These conjugated ring systems inhibit the free rotation carbon bond. Xanthones have a basic framework consisting of 13 carbon atoms that make up the composition of C6-C1-C6 (**Figure 2**).

Xanthones are compounds with the basic framework of two phenyls connected by bridges carbonyl and oxygen (ether). Their biosynthesis is not known clearly but allegedly still in close contact with the biosynthesis of flavonoids and stilbenoid. It can be seen from the type of oxygenation and two types of aromatic rings which are derived from the shikimate (shikimic acid) and the acetate-malonate pathways.

Xanthones compound that was isolated from *C. inophyllum* plants, there are prenylated and some are not prenylated. Most xanthone compounds isolated from these plants showed a characteristic, one of which is a hydroxy group at C1. The possible oxygenation position is shown in **Figure 2**.

Xanthones are known to have a variety of bioactive properties, notably the ability of antioxidants as can be seen in **Figure 3**. Mangosteen xanthones were isolated from *Garcinia mangostana* found against free radicals and prevent oxidative damage of low-density lipoprotein [16]. Moreover, isolated xanthones from mangosteen also can inhibit HL60 leukemia cells [17]. Also, α-mangosteen extracted from *G. mangostana* L. has antibacterial activity against vancomycin-resistant enterococci (VRE) and methicillin-resistant *Staphylococcus aureus* (MRSA) [18].

Various xanthone compounds can be isolated from *C. inophyllum* leaves, such as caloxanthone A, caloxanthone B, caloxanthone C, maclura xanthones, inoxanthone, calophynic acid, 3,4-dihydroxy xanthones [4, 12, 19], brasilixanthone B, buchanaxanthone [20], inophyxanthone A, pancixanthone A, gerontoxanthone B, jacareubin, pyranojacaereubin, 2-hydroxy xanthone, 4-hydroxyxanthone, 1,3,5-trihydroxy-2-methoxyxanthone, and xanthones [21, 22].

2.2 Coumarins

Coumarin (benzopyrones) compound is one of the members of benzopyrone components. In the coumarin structure, there is a benzene ring which is tied with pyrone ring [23] as can be seen in **Figure 4**. They can be divided into four main types: simple coumarins, pyranocoumarins, furanocoumarins, and

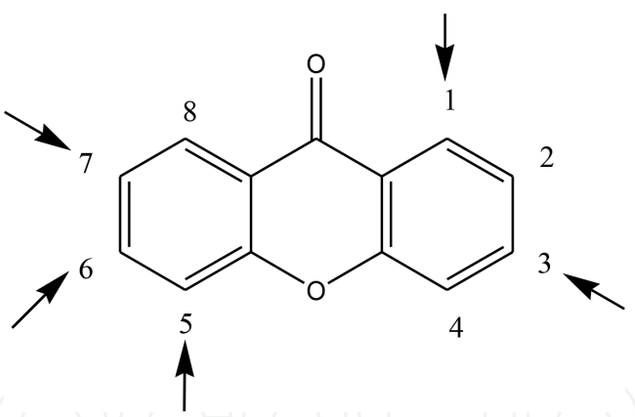

Figure 2.
Possible position oxygenation xanthone compound.

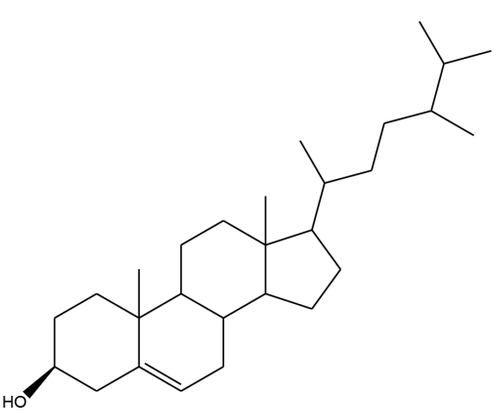

Figure 3.
Molecular structure of xanthones.

pyrone-substituted coumarins. All the reactions of coumarins focus on activation of C3,4—the double bond of the α,β-unsaturated lactone—and form a heterocyclic system [24].

Coumarins are commonly used in the agrochemical, perfume, and medical industries. They have high antitumor and antibacterial activities. Antitumor activity of 7-hydroxycoumarins against several tumor cell lines has been identified. Coumarins and their derivatives have activity as barrier against cellular proliferation in various carcinoma cell lines [25]. Besides that, they also have anticoagulant, antioxidant, antimicrobial, antiviral, anti-inflammatory, antimalarial, and analgesic activities [26].

The biosynthesis of coumarin compounds is derived from the shikimic acid pathway or still in line with the phenyl group propanoid. The skeleton benzopyran-2-on of coumarin is originating from the acid-cinnamic acid via ortho-hydrolysis. Ortho-coumaric acid produced after undergoing *cis-trans* isomerization undergoes condensation [27]. Characteristic of these compounds is their lactone group formed from the acid on the tip of propane with a hydroxy group on the phenyl group. Oxygenation coumarin compounds in the aromatic ring are also typical and are intermittent. The structure of the coumarin derivatives can be divided into four categories based on the group bound to the C_4 : 4-metilcoumarin, 4-fenilcoumarin, and 4-(n-propyl)coumarin.

2.3 Benzodipyranones

Benzodipyranones are derivative of chromone. These compounds have a skeleton similar to stilbene with two additional prenyl groups. Some benzodipyranone compounds have been isolated from the *C. inophyllum* leaves, such as (2S, 3R) and (2R, 3R)-2,3-dihydro-5-hydroxy-2,3,8,8-tetramethyl-6-(1-phenylethenyl)-4H, 8H-benzo [1,2-b: 3,4-b ‘] dipyran-4-one [14], inophynone, and isoinophynone [20, 28].

2.4 Terpenes and terpenoids

Terpenes are naturally derived component in the biosynthesis of isoprene C5 with molecular formula C_5H_8 (CH_2═C (CH_3)-CH═CH_2) (**Figure 5**). They commonly expressed in the formula $(C_5H_8)_n$ with n states the amount of isoprene which are there, so the amount of carbon is a multiple of 5. They are classified in hemiterpenes, monoterpenes (consisting of 2 units of C5 or 10 carbon atoms), sesquiterpenes (consisting of 3 units of C5 or 15 carbon atoms), diterpenes (consisting of

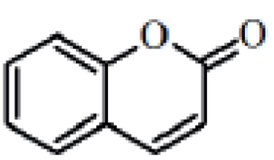

Figure 4.
Molecular structure of coumarins.

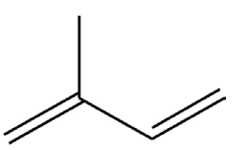

Figure 5.
Molecular structure of isoprene.

4 units of C5 or 20 carbon atoms), sesterterpenes, triterpenes (consisting of 6 units of C5 or 30 carbon atoms), tetraterpenes (consisting of 8 units of C5 or 40 carbon atoms), and polyterpenes.

Moreover, terpenoids are isoprenoid structural components which contain oxygen in its structure and can react with ketone, aldehyde, or alcohol. Chemically, they are generally soluble in fat and contained within the plant cell cytoplasm. Usually, they can be extracted with petroleum ether, ether, or chloroform and can be separated by chromatography on silica gel [29].

Terpenes are widely used as a medicine and flavor enhancers. They are commonly used in the rubber industry. They have a low molecular weight, such as essential oils that are used as natural food additives and fragrances in the perfume industry. They are also used in anticancer drug Taxol which is a diterpene. Taxol is used in the treatment of breast, ovarian, and lung cancer. One example is imberbic acid, a triterpenoid that has activity against *Mycobacterium fortuitum* and *S. aureus* [30].

Triterpenoids are a class of terpenoid compounds which consist of 30 carbon atoms or 6 units of isoprene. In plant tissue, they can be found in their native form but are also often found in the form glycoside. They are divided into cyclic and acyclic structures. The important acyclic triterpenoid is only the squalene that is considered only as an intermediate in the biosynthesis of steroids. The most widespread of triterpenoids are the pentacyclic triterpenoids. The frameworks most often found on a class of compound triterpenoids are ursam, lupan, oleanan, and friedelin [31].

Friedelin has the molecular formula $C_{30}H_{50}O$ and a molecular weight of 426,7174 g/mol (**Figure 6**). Friedelin has a melting point of 259–260°C. The structure mass spectrometry of friedelin is 426 (M^+), 411, 302, 273, 246, 231, 218, 205, 191, 179, 163, 149, 137, 125, 123, 109, 95, 81, 69, and 55. The IR spectra of friedelin in KBr was obtained using v_{max} at 1720 cm^{-1}. The form of friedelin is white crystalline-amorphous solid. Friedelin has an anti-fungal activity and has antinociceptive effects in rodents [32]. Friedelin was developed on a TLC plate by using a solvent system of 10% ethyl acetate and 90% hexane. Friedelin gave a dark spot on a TLC when exposed under UV light and iodine vapor chamber. Friedelin gave an Rf value of 0.75 with the use of a relatively nonpolar solvent system [33].

Several studies have been conducted on the benefits of friedelin. Friedelin has hepatoprotective activity [34]. It has an activity against Bacillus Calmette-Guerin (BCG) that causes tuberculosis [35]. It and some types of friedelin compound are widely used for the treatment of cancer of the bladder [36], convulsion, inflammation [37], topical ulcers, rheumatic inflammation, fever, and dysentery [38]. It is also found to have antifeedant activity in some insects [39].

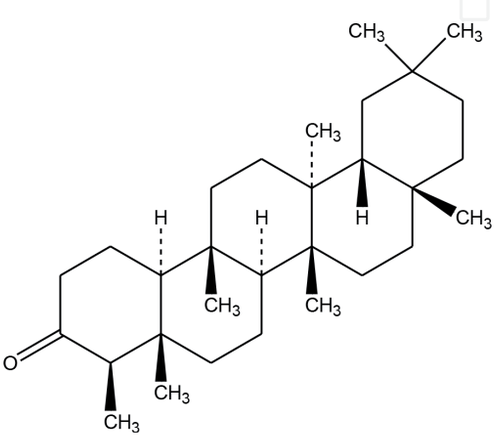

Figure 6.
Molecular structure of friedelin.

Moreover, some compound triterpenoids have been isolated from the *C. inophyllum* leaves, such as 3β, 23-epoxy-friedelane-28-OIC acid, 3-oxofriedelin-28-OIC acid, epifriedelanol, oleanolic acid [40], 3,4-secofriedelane-3,28-dioic [41], β-amyrin [20], friedelin, canophyllal, canophyllol, and canophyllic acid [4, 20, 41].

2.5 Steroid

Sterols are steroids which have a hydroxy group at C3 position as can be seen in **Figure 7**. They are found in free form or in association with glucose to form glycosides (sterolin) or as fatty acid esters (FASE). They are the natural compound that is generally composed of 27 carbon atoms [31]. They are terpenoids in which their basic framework consists of the system perhydrophenanthrene cyclopentane ring. They are a class of secondary metabolic compounds which are widely used as a drug. Steroid hormones are generally derived from natural steroid compounds, especially in plants [42]. Some steroid compounds have been isolated from the *C. inophyllum* leaves such as campesterol [20]. Campesterol also has analgesic activity.

2.6 Flavonoids

Flavonoids are the largest group of phenolic compounds found in nature, especially in tissues of higher crops. They are the product of secondary metabolites that occur from the cells and accumulate on the body crop as a toxic substance [43]. They are commonly known as flavonoids, which are water-soluble polyphenol component. They have a basic framework consisting of 15 carbon atoms where a chain of benzene (C6) is bound to a chain of propane (C3), thus forming a bond arrangement C6-C3-C6 which is particularly called phenylbenzopyran (**Figure 8**). This arrangement can produce three structures, namely, 1,3-diarilpropana (flavonoids), 1,2-diarilpropana (isoflavonoids), and 2,2-diarilpropana (neoflavonoid) [44]. Moreover, flavonoids are classified into various categories based on differences in molecular structure, such as chalcones, flavanols, catechins, flavonoes, isoflavone, dihydroflavonol, and anthocyanidins [45, 46].

According to Markham [47], flavonoids are polar compounds because they have a hydroxyl group which does not bind to sugar, so the flavonoid is quite soluble in polar solvents such as ethanol, methanol, butanol, or water. Because of the presence of sugar bound, flavonoids become more soluble in water. Conversely, the less polar aglycone, such as isoflavones, flavanones, flavones, and flavonols, which is methoxylated tends to be more soluble in solvents, such as ether and chloroform.

The largest group of flavonoids is flavones. Flavonoids have a 2-phenyl Croman order in which the ortho-position of the A ring and the carbon atom attached to the ring B of 1.3 diarilpropana is connected by bridging oxygen to form a new heterocyclic ring [47].

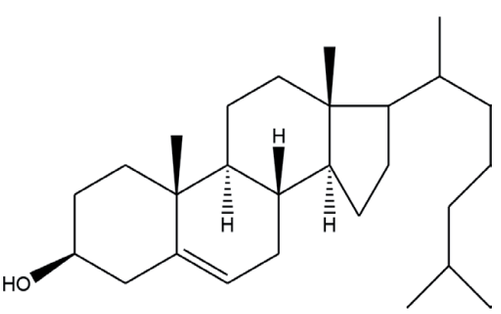

Figure 7.
Molecular structure of cholesterol.

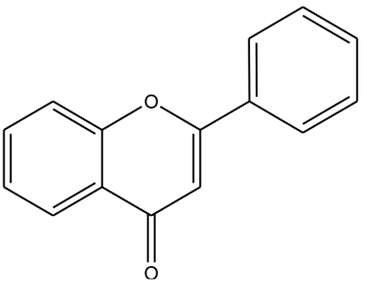

Figure 8.
Molecular structure of flavone.

Flavonoids have a variety of biological functions including pharmaceutical use and their function in plants. Examples of pigments in flowers, they provide color and attract insects for pollination. Flavonoids which are contained in the leaves have to prevent fungal infections and protect leaves from UV radiation [45]. In the aspect of pharmacology, flavonoids interact with cytochrome P450 and are used to treat heart disease. They are also known to have antioxidant activity and anti-free radicals that are useful in anticancer and antiaging. Furthermore, they also have antileukemic activity, vitamin C, 5-lipoxygenase, cyclooxygenase inhibitors, protein kinase C, tyrosine kinase, and genetic toxicity [27].

Several flavonoid compounds that have been isolated from the *C. inophyllum* leaves are bioflavonoids, neoflavonoid [48], amentoflavone [20, 40], and quercetin-3-O-α-L-rhamnoside [8, 48].

2.7 Oxygenated hydrocarbon (fatty acids)

Some of the compounds of fatty acid that has been found in the *C. inophyllum* leaves are tetradecanoic acid (myristic acid, $C_{14}H_{28}O_2$), n-hexadecanoic acid (palmitic acid, $C_{16}H_{32}O_2$), oleic acid ($C_{18}H_{34}O_2$), and octadecanoic acid (stearic acid, $C_{18}H_{36}O_2$) [49].

2.8 Esters

Some ester compounds that have been found in the *C. inophyllum* leaves are 1,2-benzenedicarboxylic acid (diisooctyl ester/phthalic acid, bis(6-methylheptyl) ester), 9,12-octadecenoic acid methyl ester, 16-octadecanoic acid methyl ester, heptadecanoic acid, and 16-methyl ester [49, 50].

2.9 Tannins

In chemistry, there are two types of tannins, namely, (1) condensed tannins or flavolan and (2) hydrolyzed tannins.

2.9.1 Condensed tannins

The condensed tannins are widespread in angiosperm plants, especially in woody plants. Another name of condensed tannins is proanthocyanidin because when they reacted with hot acid, some of the carbon-carbon connecting bond units disconnect and free monomer anthocyanidins. Most proanthocyanidin is procyanidin because when reacted with acids will produce cyanidin. Proanthocyanidin can be detected directly by dipping the plant tissue into 2 M HCl boil for half an hour that will produce a red color which can be extracted with amyl or butyl alcohol.

No.	Phytochemicals	Chemical structure	References
1.	Triterpenoids		
	3β, 23-Epoxy-friedelan-28-oic acid	$C_{30}H_{48}O_3$	[41]
	Friedelin	$C_{30}H_{50}O$	[4, 20, 28, 32, 41]
	3-Oxofriedelin-28-oic acid		[40, 41]
	Canophyllal	$C_{30}H_{48}O_2$	[20, 41]
	Canophyllol	$C_{30}H_{50}O_2$	[20, 41]
	Canophyllic acid (27-hydroxyacetate canophyllic acid)	$C_{30}H_{50}O_3$	[4, 20, 41]
	3,4-Secofriedelane-3,28-dioic acid	$C_{30}H_{50}O_4$	[19]
	Inophynone	$C_{24}H_{24}O_4$	[20, 28]
	Isoinophynone	$C_{24}H_{24}O_4$	[20, 28]
	β-Amyrin	$C_{30}H_{50}O$	[20]
	Epifriedelanol	$C_{30}H_{52}O$	[41]
	3-Oxo-27-hydroxyacetate friedelan-28-oic acid		[19]
	Oleanolic acid	$C_{30}H_{48}O_3$	[41]
	Squalene	$C_{30}H_{50}$	[50]
2.	Steroids		
	Cholesterol	$C_{27}H_{46}O$	[28]
	Campesterol	$C_{28}H_{48}O$	[20]
3.	Flavonoids		
	Biflavonoids	$C_{30}H_{20}O_{10}$	[49]
	Neoflavonoids	$C_{20}H_{18}O_8$	[49]
	Quercetin-3-O-α-L-rhamnoside (4H-1-benzopyran-4-one,2-(3,4-dihydroxyphenyl)-3,5,7-trihydroxy)	$C_{15}H_{10}O_7$	[22, 49]
	Amentoflavone	$C_{30}H_{18}O_{10}$	[20, 22, 40]
4.	Coumarins		
	Inophyllum C	$C_{25}H_{23}O_5$	[12, 40, 42]
	Inophyllum E	$C_{25}H_{22}O_5$	[12, 40]
	Inophyllum B	$C_{25}H_{24}O_5$	[4, 42]
	Inophyllum P	$C_{25}H_{24}O_5$	[4, 42]
	Calophyllic acid	$C_{25}H_{24}O_6$	[4, 20, 40]
	Isocalophyllic acid	$C_{25}H_{24}O_6$	[20, 40]
	Inophyllum G-1	$C_{25}H_{24}O_5$	[4, 42]
	Inophyllum G-2	$C_{25}H_{24}O_5$	[4, 42]
	Calocoumarin-A		[20]
	Calocoumarin-B		[20]
	Calocoumarin-C		[20]
	Apetalolide	$C_{26}H_{24}O_5$	[20]
	4-Phenylcoumarins		[20]
	Pyranocoumarins	$C_{20}H_{18}O_4$	[42]

No.	Phytochemicals	Chemical structure	References
	Calophyllolides (calophyllolide 2a, 3a, 3b, 6)	$C_{26}H_{24}O_5$	[4, 12, 42]
5.	Xanthones		
	Caloxanthone A	$C_{23}H_{22}O_6$	[4, 12]
	Caloxanthone B		[4, 12]
	Caloxanthone C		[4]
	Brasilixanthone-B	$C_{23}H_{20}O_6$	[20]
	Buchanaxanthone	$C_{14}H_{10}O_5$	[20]
	Inoxanthone	$C_{23}H_{22}O_5$	[12]
	Maclura xanthone	$C_{23}H_{22}O_6$	[12]
	Calophynic acid	$C_{35}H_{44}O_6$	[12]
	3,4-Dihydroxyxanthone	$C_{13}H_8O_4$	[12, 19]
	Inophyxanthone A		[21]
	Pancixanthone A	$C_{18}H_{16}O_5$	[21]
	Gerontoxanthone B	$C_{23}H_{22}O_6$	[21]
	Jacareubin (6-deoxyjacareubin)	$C_{18}H_{14}O_6$	[21, 22]
	Pyranojacaereubin	$C_{23}H_{20}O_6$	[21]
	2-Hydroxyxanthone	$C_{13}H_8O_3$	[22]
	4-Hydroxyxanthone	$C_{13}H_8O_3$	[22]
	1,3,5-Trihidroxy-2-methoxyxanthone		[22]
	Xanthone	$C_{13}H_8O_2$	[21]
6.	Oxygenated hydrocarbons (fatty acids)		
	Tetradecanoic acid (myristic acid)	$C_{14}H_{28}O_2$	[50]
	n-Hexadecanoic acid (palmitic acid)	$C_{16}H_{32}O_2$	[50]
	Oleic acid	$C_{18}H_{34}O_2$	[50]
	Octadecanoic acid (stearic acid)	$C_{18}H_{36}O_2$	[50]
7.	Esters		
	1,2-Benzenedicarboxylic acid (diisooctyl ester) (phthalic acid, bis(6-methylheptyl) ester) (diisooctyl phthalate)	$C_{24}H_{38}O_4$	[50]
	Methyl linoleic (9,12-octadecanoic acid methyl ester)	$C_{19}H_{34}O_2$	[50, 51]
	Methyl oleate (16-octadecanoic acid methyl ester)	$C_{19}H_{36}O_2$	[51]
	Methyl isostearate (heptadecanoic acid, 16-methyl, methyl ester)	$C_{19}H_{38}O_2$	[51]
8.	Alkenes (unsaturated compounds):		
	Azulene, 1,4-dimethyl-7-(1-methylethyl)-	$C_{15}H_{18}$	[50]
9.	Ethers		
	3-Trifluoroacetoxypentadecane (pentadecyl trifluoroacetate) (trifluoroacetic, pentadecyl ester)	$C_{17}H_{31}F_3O_2$	[50]
	1-Monolinoleoglycerol trimethylsilyl ether	$C_{27}H_{54}O_4Si_2$	[50]
10.	Alicyclic compounds		
	Cyclohexene, 3-(1,5-dimethyl-4-hexenyl)-6-methylene-,[S-(R*,S*)]	$C_{15}H_{24}$	[50]

No.	Phytochemicals	Chemical structure	References
11.	Aromatic hydrocarbon:		
	Benzene (1-methyldodecyl)	$C_{19}H_{32}$	[50]
12.	Androstan-1α-ol-17-one,23 isopropylidenedioxy-4β-methyl-	$C_{23}H_{36}O_4$	[50]
13.	Proanthocyanidin (condensed tannin)	$C_{31}H_{28}O_{12}$	[20, 49]
14.	Benzodipyranone (chromone) derivatives:		
	a. (2S,3R)-2,3-Dihydro-5-hidroxy-2,3,8,8-tetramethyl-6-(1-phenylethenyl)-4H,8H-benzo [1,2-b:3,4-b'] dipyran-4-one		[14]
	b. (2R,3R)-2,3-Dihydro-5-hidroxy-2,3,8,8-tetramethyl-6-(1-phenylethenyl)-4H,8H-benzo [1,2-b:3,4-b'] dipyran-4-one		[14]
15.	Asam inophylloidic	$C_{32}H_{46}O_6$	[12, 20]
16.	Calaustralin	$C_{25}H_{25}O_5$	[12]
17.	Shikimic acid	$C_7H_{10}O_5$	[40]
18.	Brasiliensic acid	$C_{32}H_{46}O_6$	[12]
19.	Adenanthin (7,8,12-tri-0-acetyl-3-desoxy-ingol3-one)	$C_{26}H_{34}O_9$	[51]
20.	Carbazole	$C_{12}H_9N$	[51]
21.	Diphenyl methane (1'-biphenyl, 2-methyl)	$C_{13}H_{12}$	[51]
22.	2-Phenazinamine (1,1'-biphenyl, 4-azido)	$C_{12}H_9N_3$	[51]
23.	5-Aminomethyl-dibenzosuberane (2-naphtalenecarbonitrile, 6-pentyl-)	$C_{16}H_{17}N$	[51]
24.	Phytol	$C_{20}H_{40}O$	[50, 51]
25	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	$C_{20}H_{40}O$	[50]
26.	Phenol (2,4-bis(1-phenylethyl)-phenol)	$C_{22}H_{22}O$	[50]

Table 2.
Phytochemicals contained in the C. inophyllum leaves.

When dry tissues are used, the result of tannins somewhat diminished because of the occurrence of sticking tannins in place within the cell.

2.9.2 Hydrolyzed tannins

The hydrolyzed tannins are contained in dicotyledonous plants. They mainly consist of two classes; the simplest is galloylglucose. In this compound, glucose is surrounded by five or more galloyl ester groups. The second type is the core molecules of a compound gallic acid dimer, namely, hexahydroxidifenate acid that binds to glucose. Hydrolyzed tannins can be detected by determining the gallic acid or ellagic acid in ether or ethyl acetate extracts.

2.10 Other components

Some chemical compounds that have been found in the *C. inophyllum* leaves are azulene ($C_{15}H_{18}$), squalene ($C_{30}H_{50}$), 3-trifluoroacetyl pentadecane (pentadecyl trifluoroacetate), 1-monolinolein glycerol trimethylsilyl ether, cyclohexane, benzene, androstane [49], inophylloidic acid [12, 20, 52], shikimic acid [40], calaustralin, brasiliensic acid [12], adenanthin, carbazole, diphenyl methane, 2-phenazinamine, 5-aminomethyl-dibenzosuberane [50], phytol, phenol, and 3,7,11,15-tetramethyl-2-hexadecene-1-ol [49, 50]. The summary of phytochemicals in *C. inophyllum* leaves is presented in **Table 2**.

3. Isolation method of phytochemicals in *C. inophyllum* leaves

Polarity is one of the characteristics of chemical bonding, where two different atoms within the same molecule have a different electronegativity. As a result, the electrons in the bond are not shared equally by the two atoms. This causes the electric field (pole) to be asymmetric. Covalent bonding of molecules can be described as polar or nonpolar.

The polar compound is a compound formed by a single atom which has electronegativity substantially greater than the other. The more electronegative the atom, the pull of the bonding electrons is greater. The result is a bond with an uneven electron dense distribution. The nonpolar compound is a compound formed by atoms with the same or nearly the same electronegativity and forms covalent bonds, where both atoms apply traction which equals or nearly equals to the bonding electrons. Generally, the carbon-carbon and carbon-hydrogen bonds are the most common types of nonpolar bond [53].

To identify polar and nonpolar compounds from the *C. inophyllum* leaves, the first idea is separating their compounds based on the solvent used (solvent polarity index). Methanol and water are polar solvent with a polarity index of 5.1 and 9, respectively. For n-hexane or petroleum ether is nonpolar solvent with a polarity index of 0 [54]. It can be expected that polar compounds which are contained in the *C. inophyllum* leaves can be dissolved in a polar solvent and vice versa. Relative polarities of several solvents can be seen in **Table 3.**

Extraction is the separation process of material from a solid or some material from liquid with the help of the solvent. Extraction can be defined as a method of separating components of a mixture by using a suitable solvent. Solutes (dissolved substances) are separated in a manner distributed between two layers of solvents based on their solubility. Extraction is a separation of the compounds contained in the liquid material/solid using certain solvents at any given temperature.

In general, extraction techniques can be classified into two general categories:

1. Short-term extraction is extraction techniques typically used to separate a substance (liquid form), on the basis of differences in solubility of the two immiscible solvents.

2. Long-term extraction is an extraction technique normally used to separate the natural material (solid form) contained in plants or animals. It is a classic procedure to obtain the organic matter content of dry plant tissue by soaking with certain solvents (polar or nonpolar solvents) [29].

Percolation is an extraction technique that done repeatedly and performed at a room temperature. This is similar to maceration, but after soaking for a certain time, the solvent is removed and replaced with a new solvent. After filtration, the filtrate obtained is called percolate [55].

According to Mulyono [55], in terms of the extraction mechanism, known to some type of extraction, namely:

1. Single-stage extraction

Single-stage extraction is the extraction method using a single type of solvent, and extraction is only done once with a solvent.

Relative polarity	Formula	Group	Solvents
Nonpolar	R-H	Alkanes	Petroleum ethers, hexanes, ligroin
	Ar-H	Aromatics	Toluene
	R-O-R	Ethers	Diethyl ether
	R-X	Alkyl halides	Trichloromethane, chloroform
	R-COOR	Esters	Ethyl acetate
	R-CO-R	Aldehydes, ketones	Acetone, MEK
	R-NH2	Amines	Pyridine, triethylamine
	R-OH	Alcohols	MeOH, EtOH, IPA, butanol
	R-COHN2	Amides	Dimethylformamide
	R-COOH	Carboxylic acid	Ethanoic acid
Polar	H-O-H	Water	

Table 3.
Relative polarity of solvents [54].

2. Repeated extraction

Repeated extraction is the extraction method using a solvent, but the process is repeated with a number of solvents.

3. Stage extraction

Stage extraction is the extraction method using some type of solvent extraction, such as after extraction with the first solvent, followed by using other solvents, and so on.

Solvents are not or only partially soluble solids or liquids with continuous contact; the active agents move from a mixture of solids/liquid (raffinate) to the solvent (extract). After mixing the two phases, the separation process is done on the principle of gravity or centrifugal force [56].

Yunitasari [57] describes the effect of solvent on the various types of tray number from 6 to 10 for taking *C. inophyllum* oil with column extraction. From the experimental results, the authors explain that the more the number of trays, the less time is required for a solvent to extract the oil. The solvent used are between n-petroleum and n-hexane. From the experimental results, the authors explain that the maximum condition extraction was achieved by n-petroleum in the seventh tray. The amount of oil was decreasing by increasing number of tray. In the other hand, the amount of oil was increasing with number of tray while n-hexane was used.

4. Conclusions

The identification and uses of beneficial phytochemicals contained in *C. inophyllum* leaves were presented in this book chapter. It was found that all parts of *C. inophyllum* plant can be used for human needs. The information is limited to extraction and identification of mixture of phytochemical compounds that are obtained from plant extracts. The separation of individual phytochemical compounds still remains unknown. Therefore, further research on the determining of phytochemicals content in this plant is necessary.

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Conflict of interest

We declare that we have no conflict of interest.

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