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Recent Advances on Pharmacology and Chemistry of *Pycnanthus angolensis* over the last decade (2012-2021)

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ABSTRACT: *Pycnanthus angolensis*, widely known under its trade name "ilomba", is a medicinal plant from the family Myristicaceae that has occupied a prominent place in African traditional medicine for several decades; its broad application to treat numerous diseases, including malaria, bacterial infections and most recently COVID-19. The various chemical studies undertaken on the plant identified many classes of specialized compounds, including quinone-terpenoids, lignans and isoflavonoids, as the most abundant and bioactive components. The plant is defined as a major asset in developing new potent drugs and deserves further investigation in this regard. This mini-review aims to compile the newly documented findings on the traditional uses, phytochemistry and pharmacology of *P. angolensis* over the last decade from 2012 to 2021. In this regard, a literature search using the keyword *Pycnanthus* has been done without language restriction in numerous online libraries, including Scifinder, PubMed, Google Scholar, and only papers on *Pycnanthus angolensis* published after 2011 have been exploited during the writing.

1. INTRODUCTION

It is well-known today that medicinal plants occupy a prominent place in the mechanisms of treatment of various illnesses in traditional medicine and therefore represent an important source of production of the active ingredients which attract the attention of researchers and the pharmaceutical industries in the development of new effective drugs against various diseases (Happi et al., 2022). Therefore, it is not surprising that traditional medicine is an important part of health care in many countries of the world. More specifically, this medicine relates to health practices and knowledge incorporating herbal medicines to treat or prevent disease (Fokunang et al., 2011). During the COVID-19 pandemic, great attention has been given to developing several improved traditional medicines from medicinal plants to control and heal the infected persons, mostly in developing countries waiting for approved drugs, vaccines or effective treatment (Fedoung et al., 2021; Panchamoorthy & Vel, 2022). *Pycnanthus angolensis* is a well-known plant in African traditional medicine for its general use in treating several diseases from West Africa to Tanzania. It has been long investigated for its chemical constituents, pharmacological activities and, more recently, its traditional use in the treatment of COVID-19, for example (Fedoung et al., 2021). In 2012, a first review summarizing the initial investigations on the plant up to 2011 was published (Achel et al., 2012). During the last decades, additional research works have been globally done on the plant and newly reported specialized metabolites, traditional uses and pharmacological data have been documented. This paper aims to compile all these recent data published on *P. angolensis* over the last decades from 2012 to 2021.

2. BOTANY AND DISTRIBUTION OF *PYCNANTHUS ANGOLENSIS*

The genus *Pycnanthus* has three accepted species from the Myristicaceae family, all native to tropical Africa, including *P. dinklagei* Warb, *P. marchalianus* Ghesq and *P. angolensis* (Welw.) Warb is the most encountered and exploited Theplantlist (2021). More significantly, *P. angolensis* also called "false nutmeg", "African nutmeg", "ilomba" (trade name) or "mulomba" in Gabon, which means "holy tree", is a tree distributed from West Africa to Tanzania crossing by Nigeria, Cameroon, Gabon and DR Congo. The tree is about 25 to 40 m high and 60 to 150 cm in diameter with a straight and cylindrical bole. The wood varies from a greyish white to pink colour, and the branches are in whorls while the leaves are generally 18 to 31 cm in length and 5 to 9 cm in width. They have almost parallel margins, acuminate at the apex, cordate at

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the base. The leaf's midrib is very prominent below, with the lateral part about 1.3 cm long. The flowers are densely clustered at the ends of irregularly branched panicles 7.5 to 15 cm long, erect among the leaves or slightly below them. The stamens are visible after the opening of the male flowers. The fruits (often present in the flowers) are ellipsoid or almost spherical drupes, 2.5 to 3.8 cm long and 1.9 to 3.2 cm in diameter, often in dense clusters at the base of the twigs, opening with two valves and exposing a solitary black seed with a bright red aril very branched at the apex (Orwa et al., 2009). Figure 1 shows the photographs of the trunk, leaves and fruits of *P. angolensis*.

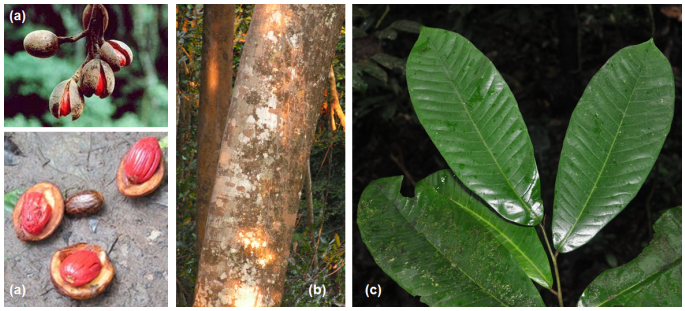

Figure 1. Photographs of *P. angolensis*. (a) : fruits, (b): trunk, (c): leaves.

3. ETHNOBOTANY OF *PYCNANTHUS ANGOLENSIS*

Pycnanthus angolensis is an essential medicinal tropical tree species with multiple therapeutic values reported in the literature. The local population widely uses the plant as the first line in treating several diseases in folk medicine. In Gabon, Baghama and Babongo tribes use the stem barks of the plant for protection against wizarding attacks (Kwenzi-Mikala & Mbadinga, 2009). In the Democratic Republic of Congo, the decoction of the bark of the trunk of *Pycnanthus angolensis* associated with the root of *Elaeis guineensis* help in the management of cough, fever and anaemia. In contrast, the juice from the trunk bark associated with salt is used to treat dental caries, and the decoction of the root is taken one glass twice a day for four days as treatment of gastritis (Mbuta et al., 2012).

Furthermore, our survey indicates that in Nigeria, the twigs of *Pycnanthus angolensis* are chewed for dental hygiene and against oral candidiasis (Kayode & Omotoyinbo, 2009), whereas the maceration of stem bark is prescribed in case of tuberculosis (Ogbole & Ajaiyeoba, 2010). In case of dysentery, fever or anaemia, the decoction of bark and leaves of *P. angolensis* is orally taken in Cameroonian folk medicine (Jiofack et al., 2009). Additional reports in the literature indicated that the plant is also used as an antidote and in the treatment of fungal infections, dermatoses, digestive problems, malaria, as well as to facilitate birth (Bene et al., 2019; Fedoung et al., 2021; Ohashi et al., 2018; Reddel et al., 2014) .

4. RECENT COMPOUNDS REPORTED FROM *PYCNANTHUS ANGOLENSIS* AND THEIR ACTIVITY

A decade later, after the systematic review on *Pycnanthus angolensis* published by Achel et al. (2012) , the literature survey indicates that only nineteen additional compounds, including seven lignans (**1**–**7**), seven isoflavonoids (**8**– **14**), one flavonoid (**15**), one diterpenoid (**16**), two acid derivatives (**17**– **18**) and one fluorene (**19**) have been isolated from different organs (stem bark, seed, fruits, root or leaves) of the plant (Table 1).

Table 1
Compounds isolated from *P.angolensis* over the last decade (2012-2021).

Classes	N°	Name	Organ	References
Lignans	**1**	Talaumidin	Stem bark	Mansoor et al. (2012)
	2	Hinokinin		
	3	Threo-4,4'-dihydroxy-3-methoxylignan		
	4	4'-Hydroxy-3,3',4-trimethoxylignan		
	5	(-)-Dihydroguaiaretic acid		
	6	3,3',4,4'-tetramethoxylignan		
	7	4,4'-Diacetyl-3,3'-dimethoxylignan		
Flavonoids	**8**	4',7-Dihydroxy-2'-methoxyisoflavan	Stem bark	Mansoor et al. (2011)
	9	Calycosin	Fruits	
	10	Formononetin	Root	
	11	2'-Hydroxybiochanin A	Stem bark	
	12	Irilone		
	13	Tectorigenine		
	14	Genistein		
	15	Liguiritigentin		
Miscellaneous	**16**	Eluptol	Leaves	Elufioye et al. (2016)
	17	Omifoate A		
	18	1,6-Dihydro-2-methyl-4-hydroxy-6-oxo-3-pyridine carboxylic acid, ethyl ester	Seed	O.J. Oladimeji and Onu (2018)
	19	9-oximino-2,7-diethoxy fluorene	Leaves	O.H. Oladimeji and Ahmadu (2019)

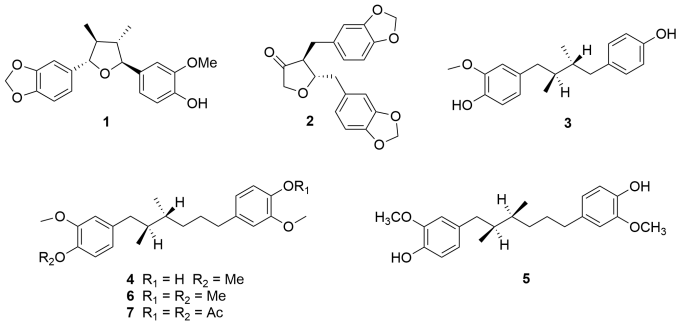

Figure 2. Lignans (**1**– **7**) isolated from *P. angolensis*.

Lignans are a significant class of compounds isolated in the family Myristicaceae with isoflavonoids. Seven lignans (Figure 2), namely talaumidin (**1**), hinokinin (**2**), threo-4,4'-dihydroxy-3-methoxylignan (**3**), 4'-hydroxy-3,3',4-trimethoxylignan (**4**), (-)-dihydroguaiaretic acid (**5**), 3,3',4,4'-tetramethoxylignan (**6**), 4,4'-diacetyl-3,3'-dimethoxylignan (**7**) were isolated from the stem bark of *P. angolensis* and tested for their ability as apoptosis inducers in human hepatoma HuH-7 cells. The results indicated that the compounds **1**-**7** displayed a significant activity by reducing the viability of HuH-7 cells and, more significantly, hinokinin (**2**) and 4'-hydroxy-3,3',4-trimethoxylignan (**4**) were the most potent inducers with 2.5- and 2.4-fold increases in apoptotic cells as compared to controls (Mansoor et al., 2012).

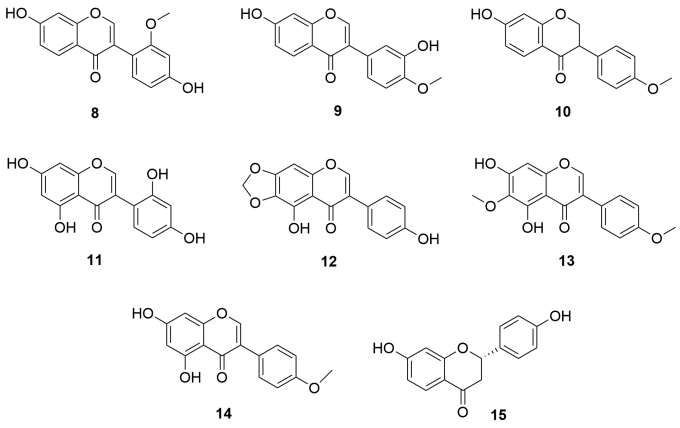

Figure 3. Isoflavonoids (**8**– **14**) and flavonoid (**15**) isolated from *P. angolensis.*

A similar study was done by Mansoor et al. (2011) on isoflavonoids and flavonoids isolated from the stem bark of *P. angolensis*. Briefly, the recently reported isoflavonoids 4′,7-dihydroxy-2′-methoxy-isoflavan (**8**), calycosin (**9**), formononetin (**10**), 2'-hydroxybiochanin A (**11**), irilone (**12**), tectorigenine (**13**), genistein (**14**) and liguiritigentin (**15**) (Figure 3) have also been evaluated for their ability as apoptosis inducers in human hepatoma HuH-7 cells. As expected for flavonoids that are significant apoptosis inducers due to their antioxidant character and good interaction with enzymes, all the tested flavonoids displayed significant apoptosis induction. Therefore, irilone (**12**) demonstrated the highest activity reflecting an increase of almost 80 % in caspase-3 while 4',7-dihydroxy-2'-methoxy-isoflavan (**8**), formononetin (**10**), tectorigenine (**13**) and genistein (**14**) gave at least a 50 % increase in caspase activity (Mansoor et al., 2011).

A comprehensive review dedicated to the anticancer potency of formononetin (**10**) and its action mechanisms has been documented in the literature (Jiang et al., 2019). At least sixteen plants were identified as sources of formononetin in this study, including *P. angolensis*, whose bark yielded 0.009 % of this lead component. Formononetin (**10**) shows eight distinct bioactivity categories, including anti-inflammatory, antioxidant, vasodilator, antibacterial, antiallergic, neuroprotective, cardioprotective, and anticancer properties. Based on the previous reports on *in vitro* and *in vivo* anticancer as well as biosafety profile of formononetin (**10**), the compound was qualified as a good and safe candidate for new promising and active drug development for several illnesses like nasopharyngeal carcinomas, for example, since the compound displayed an intense activity (IC_{50} = 1 μM) against the CNE2 cell lines. However, despite its good apoptosis induction activity in several tumour cells, its action mechanism deserves further investigations to be established (Jiang et al., 2019).

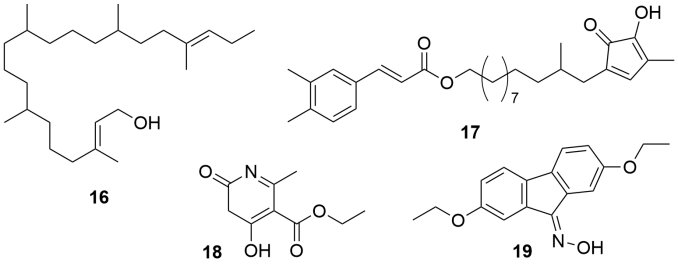

Figure 4. Miscellaneous compounds isolated from *P. angolensis*

Finally, four other miscellaneous compounds (Figure 4) have been reported during the phytochemical study of leaves, seeds and roots of *P. angolensis*. The acyclic diterpenoid eluptol (**16**) and the cinnamic acid ester derivative omifoate A (**17**) were obtained from the leaves of *P. angolensis* and evaluated for their acetylcholinesterase (AchE) and butyrylcholinesterase (BuChE) inhibitory activities. Both compounds showed good activities with IC_{50} values of 22.26 μg/ml (AChE) and 34.61 μg/ml (BuChE) for compound **16** and 6.51 μg/ml (AChE) and 9.07 μg/ml (BuChE) for compound **17** (Elufioye et al., 2016). Furthermore, 1,6-dihydro-2-methyl-4-hydroxy-6-oxo-3-pyridine carboxylic acid, ethyl ester (**18**) was evaluated by well-diffusion against two bacterial strains, *Staphylococcus aureus* (ATCC 21824) (Gram-positive) and *Escherichia coli* (ATCC 23523) (Gram-negative), as well as one fungal strain *Candida albicans* (NCYC 106). The results showed that compound **18** was bacteriostatic against *E. coli* with a zone of inhibition of 11.5 mm at 2 mg/ml and was not active against the other microbial strains (O.J. Oladimeji & Onu, 2018). The last compound, 9-oximino-2,7-diethoxyfluorene (**19**), was reported from *P. angolensis* leaves and demonstrated a radical scavenging activity with an IC_{50} value of 0.58 μg/ml (Adu et al., 2019).

5. BIOLOGICAL EVALUATIONS OF *PYCNANTHUS ANGOLENSIS* EXTRACTS

A great variety of biological activities performed on the *P. angolensis* extracts and compounds has been compiled in the first review published on that plant species (Achel et al., 2012). These various activities supported the importance and the use of the plant in traditional medicine. Beyond this first batch of pharmacological tests, researchers have continued to evaluate the extracts of the plants during the last decade and reported additional results that enrich the plant's pharmacology. The methanol soluble extract *P. angolensis* stem bark showed the

same antibacterial activity against *S. aureus* and *B. subtilis* with the same IC_{50} values of 14.6 mg/ml and 14.7 mg/ml (Adu et al., 2019). Bosson-Vanga et al. (2018) reported that the same methanolic extract of *P. angolensis* stem bark displayed a weak antifungal against *C. albicans* with IC_{50} values ranging from 12.5 to 100 mg/ml.

Further analyses of *P. angolensis* extracts in cytotoxicity activity against the human cervix adenocarcinoma cell lines by resazurin reduction indicated that the dichloromethane soluble stem bark extract displayed higher cytotoxicity with CC_{50} of 26.66 μg/ml. In comparison, the ethyl acetate soluble extract demonstrated significant cytotoxicity with CC_{50} of 90.27 μg/ml (Okoro & Tor-Anyiin, 2017). The works of Elufioye et al. (2016) reported that the ethyl acetate soluble extract of *P. angolensis* leaves strongly inhibited acetylcholinesterase and butyrylcholinesterase enzymes at 65.66 $\pm$ 1.06 % and 49.38 $\pm$ 1.66 %, respectively. Recent works of Achel et al. (2020) reported the first evaluation *in vitro* and *in vivo* of the antimutagenic and radioprotective activities of the seed extract of *P. angolensis*. Their results established that the extract displayed a strong genoprotective ability against X-rays supported by a 35% (protection factor) rate in the two models of tests.

Additionally, they reported that the same seed extract of *P. angolensis* can eliminate up to 43.8% (equal to a protection factor of 35.5%) of X-ray-induced cell death in the radiosensitive cells while it showed only a protection factor of 29% in the radioresistant cells. These results supported that the *P. angolensis* seed extract is an essential candidate in developing neutraceutical radioprotectants. The dichloromethane soluble extract of *P. angolensis* stem bark demonstrated relevant memory-enhancing activity in mice by increasing its alternation behaviour in the Y-maze and reducing its escape latency and transfer latency in the Morris water maze paradigm and the elevated plus-maze, respectively. This result was further biochemically supported by the established evidence about the action of the dichloromethane extract of *P. angolensis* in the mice brain by increasing, on one hand, glutathione and superoxide dismutase and, on the other hand, decreasing malondialdehyde and AChE activity (Patricia et al., 2019).

6. CONCLUSION

This mini-review follows from the first one published in 2012 on the medicinal properties and applications of the plant *P. angolensis* widely used in traditional African medicine. Our research in the literature shows that nineteen compounds have been newly isolated from the plant over the past decade and tested in various biological activities. More significantly, the lignans hinokinin (**2**) and 4'-hydroxy-3,3',4-trimethoxylignan (**4**) as well as the flavonoids irilone (**12**), 4',7-dihydroxy-2'-methoxy-isoflavan (**8**), formononetin (**10**), tectorigenine (**13**) and genistein (**14**) demonstrated intense apoptosis induction activities in human hepatoma HuH-7 cells. We also found that extracts and isolated compounds like 1,6-dihydro-2-methyl-4-hydroxy-6-oxo-3-pyridine carboxylic acid, ethyl ester (**18**) were bacteriostatic against bacterial strains of *S. aureus* and *B. subtilis* but not active against the fungal strain *C. albicans*. This result might indicate that the plant is not appropriate for fungal infections, but further analyses are necessary to establish a confident conclusion in this regard. Additional uses of the plant in pharmacopoeia and new biological tests on the reported extracts are also summarized. Besides contributing to the collection of information on this medicinal plant of great importance and supporting its use in traditional medicine, this article enriches the literature on the phytochemistry and pharmacology of *P. angolensis* in order to bring and make available the information from previous work to readers and researchers for further work in continuity.

CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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AUTHOR CONTRIBUTIONS

G.M.H. designed the project, collected the data and wrote the paper. J.D.W. carried out the critical revision and the final approval of the paper. K.G.S and L.C.D equally contributed to the work by analysing and interpreting the data and drawing the structures.

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