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Phytochemical Analyses of the Menthanol, Hot Water and N-Hexane Extracts of the Aerial Parts of *Cassytha Filiformis (Linn)* and Leaves of *Cleistopholis Patens (Benth)*

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ABSTRACT

Phytochemical studies of the aerial parts of *Cassytha Filiformis Linn* and leaves of *Cleistopholis patens Benth* were analysed. The phytochemical analyses of the methanol, hot water and n-hexane extracts of *Cassytha Filiformis Linn* and *Cleistopholis patens* showed the presence of glycosides, steroids, terpenoids, alkaloids, carbohydrates, acidic compounds, resins, saponins, tannins, fats and oil and flavonoids. Phytochemical like proteins was not detected.

Keywords: *Cassytha Filiformis Linn, Cleistopholis patens Benth,* Phytochemicals, Flavonoids and acidic compounds.

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INTRODUCTION

People on all continents have used hundreds to thousands of indigenous plants for the treatment of ailments since pre-historical time. Spices and herbs are generally known to posses antibacterial and antioxidant properties [20]. Many medicinal plants are used in modern medicine where they occupy a very significance place as raw material for important drugs and plants [12]. Medicinal plants are of great importance to the health of individual and communities. Phytochemicals are chemical compounds that are naturally found in plant. They are responsible for the colour and organoleptic properties of the plant. Phytochemicals could be available as dietary supplements, but the potential health benefits of phytochemicals are derived from consumption of the whole plant [13].

Cassytha filiformis is a leafless, perennial, climbing, twinning, vine-like, authoparasitic and plant- hyperparasitic phanerogram (seed-bearing plant) in the plant family *Lauraceae.* The stems, which are green to orange, filiform and glabrous are long and thread like, measuring 3-8m in length, grows in a triangle on hosts. Flowers are sessile and few in spicate inflorescence 1-2cm long. *Cassytha filiformis Linn* inhabits coastal vegetation, usually on sandy dunes, margins of evergreen gully and venerin forests, deciduous and coastal woodlands, montane grassland and dambo margins and vegetation of sandy beaches or lakes. It grows on small bushes, woods shrubs and low tree, grasses and herbaceous weeds [15]. Its seeds might be dispersed by animals, man, water, strong winds, farm machinery. They parasitize other plants by specialized attachment and penetration with feeding structures known as *haustoria.* According to [16], "this plant is distributed throughout India and is used for medicinal purposes in China, Madgascar and South Africa.

In medicine, it is used traditionally for treatment of some human birthing issues. Modern midwives recommend taking the juice made from crushed vines for 4 weeks before the expected date of birth in order to ease labour pains and to quicken labour time and lubricate the birth canal [9]. *Cassytha filiformis* is also purported to be used by several different Polynesian cultures for the treatment of cancers. In modern medical research, *Cassytha filiformis* has a number of biologically active chemical compounds with potential human health application. Ocoteine isolated from *Cassytha filiformis,* is an alpha-adrenoreceptor antagonist in rat thoracic aorta, have antiplelet aggregation activity [1]. *Cassytha filiformis* is used as vasorelaxant [20] and adrenoreceptor antagonist [1], antitrypanosomal agent [1] and diuretics in traditional medical practice [8].

Cleistopholis patens (Benth) is a tree 20-30m tall, with a trunk up to 10m tall and 20-90cm wide, its back is grayish-white, smooth fibrous or furrowed . It is a fast growing, commonly seen in forests and rapidly colonizing abandoned areas. The bole is slender, cylindrical and straight and its timber is straight grained. The tree is sun-loving, common in distributed forest and rapidly colonizing abandoned areas. It is used in traditional medical practices in many parts of Africa where it has several applications. [6] isolated oligorhamnosides anti –bacteria from *Cleistopholis patens Benth.* In addition, alkaloids, (eupolauridine) and 3-methoxy champangine, from the ethanolic extract of *Cleistopholis patens*

Benth are potent anti-fungal agents [10] reported that the steroidal, glycoside and alkaloidal fractions of *Cleistopholis patens Benth* are effective against *klebsiella pneumonia.*

This study was designed to determine the phytochemical constituents of hot water, methanol and n-hexane extracts of the aerial parts of *Cassytha Filiformis Linn* and leaves of *Cleistopholis patens Benth*.

MATERIALS AND METHODS

Plant material

Fresh aerial parts of *Cassytha Filiformis Linn* and leaves of *Cleistopholis patens Benth* were collected from Nsukka in Enugu State Nigeria between June to August, 2009. The aerial parts and leaves were indentified by Mr. A.O Ozioko of the Bioresource development and conservation programme (BDCP) Nsukka, Enugu State, Nigeria.

The two plants were dried under the shade for 10 days, pulverized and stored at room temperature for two weeks before extraction processes.

Chemicals/Reagents

All chemicals used in this study were of analytical grade and products of May and Baker, England; BDH, England and Merck, Darmstand, Germany.

Extraction Procedure

The fresh aerial parts of *Cassytha Filiformis Linn* and leaves of *Cleistopholis patens Benth* plant were plucked and shade dried for 10 days, after which the two plants were pulverized into coarse form with a crestor high speed milling machine and stored for two weeks. Approximately, 400g each of the coarse forms of *Cleistopholis patens Benth* and *Cassytha Filiformis Linn* were then macerated in 2.0L and 4.0L of hot water respectively and allowed to stand at room temperature. This was left to stand for 48 hours. After that the hot water extracts were filtered through muslin cloth on a plug of glass wool in a glass column. The resulting hot water extracts were concentrated and evaporated to dryness using rotary evaporator at an optimum temperature of between 40 and 45^{o}C to avoid denaturation of the active ingredients. The concentrated extracts were then stored in the refrigerator. The methanol and n-hexane extracts of the two plants were obtained by soxhlet extraction. Approximately, 300g of pulverized *Cassytha Filiformis Linn* and 400g of pulverized leaves of *Cleistopholis patens Benth* were extracted with 1.5L and 1.6L of menthanol respectively. Finally, 400g of pulverized *Cassytha Filiformis Linn* was extracted with 1.5L n-hexane.

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Determination of yield of extracts

The percentage yield of the extracts were determined by weighing the coarse *Cassytha Filiformis Linn* and *Cleistopholis patens Benth* before extraction and *Cassytha Filiformis Linn* and leaves of *Cleistopholis patens Benth* extracts after concentration and then calculated using the formula.

$$\text{Percentage (\%) yield} = \frac{\text{Weight (g) of the concentrated extracts}}{\text{Weight (g) of the ground plant extracts}} \text{x100}$$

Determination of phytochemicals

The qualitative phytochemical analyses of the crude hot water, methanol and n-hexane extracts of both plants' parts were carried out based on the procedure outlined [5] and [19].

RESULTS

Percentage yield of the extracts

The percentage yield of all extracts of *Cleistopholis patens Benth* were greater than that for *Cassytha Filiformis Linn.* The yield per plant extract increased with increasing polarity of the solvent; highest yield was noted with hot water, followed by methanol and the least was n-hexane extract.

Table 1: Percentage yield of the extracts of aerial parts of *Cassytha Filiformis Linn* and *Cleistopholis patens Benth*

Plant	Part	Percentage yield of the extracts (%)	Percentage yield of the extracts (%)	Percentage yield of the extracts (%)
		Hot water	Methanol	n-hexane
Cassytha Filiformis Linn.	Aeriel parts	12.5	5.8	12
Cleistopholis patens Benth	Leaves	12.9	10.5	-

The results from Table 2: shows the presence of glycosides, steroids, terpenoids, alkaloids, carbohydrates, acidic compounds, resins, saponins, tannins, fats and oil and flavonoids. Phytochemical like proteins was not detected.

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Table 2: Phytochemical composition of the extracts of aerial parts of Cassytha Filiformis Linn and Cleistopholis patens Benth .

Phytoconstituents	Cassytha Filiformis Linn			Cleistopholis patens Benth		
	MF	AF	NF	MP	AP	NP
Glycosides	+++	++++	-	++	+	-
Steroids	-	-	++	+	-	+
Terpenoids	+	-	++	+++	-	-
Alkaloids	+	+++	-	+	-	-
Acidic compounds	-	+	-	-	-	-
Carbohydrates	+	++	-	++	+	-
Resins	-	-	+++	-	+	++
Tannins	++	+++	-	-	+	-
Saponins	-	-	-	+++	+	-
Flavonoids	+	-	++	++	-	-
Proteins	-	-	-	-	-	-
Fat and oils	-	-	+	++	-	+

Key to abbrevations
MF: Methanol extracts of Cassytha Filiformis Linn aerial parts
AF:Hot water extracts of Cassytha Filiformis Linn aerial parts
NF: n-Hexane extracts of Cassytha Filiformis Linn aerial parts

MP: Methanol extracts of Cleistopholis patens Benth leaf
AP: Hot water extracts of Cleistopholis patens Benth leaf
NP: n-Hexane extracts of Cleistopholis patens Benth leaf

++++ : Abundantly present
+++: Copiously present
++ : Moderately present
+ : Slightly present
- : Absent

DISCUSSION

The phytochemical analysis of the aerial parts of *Cassytha Filiformis Linn* revealed the presence of flavonoids, alkaloids, glycosides, terpenoids, steroids, fats and oil, carbohydrates, resins and tannins, but proteins and saponins were found to be absent in the extract. In the methanol extracts of the same plants glycosides were

Copiously present whereas other constituents occurred in moderate or slight amounts. While, in hot water extracts, glycosides occurred at abundant level, alkaloids and tannins at copious amounts and carbohydrates at low level. Steroids, terpenoids, flavonoids and reducing sugars exist at substantial amounts in n-hexane fraction of the plant. These metabolites have been shown to be responsible for various therapeutic activities of medicinal plants [19]. For instance, flavonoids acts as a “biological response modifiers” such as anti-allergic, anti-inflammatory, ant-microbial [3], and anti-cancer activities shown from *in vitro* studies [11], [14]. According to [2] flavonoids are especially known for their anti-fungal effects against wide array

of micro-organisms, the activities are attributed to their abilities to complex with extracellular and soluble proteins with microbial cell wall.

Tannins have been found to form irreversible complexes with proline proteins resulting in inhibition of cell protein synthesis. Tannins are potent antioxidants and also used for treating diarrhea and dysentery. Alkaloids, the largest group of chemicals, produced by plants have many biological activities. Alkaloids (pure and synthetic) isolates are used as basic medicinal agents for their analgesic, antispasmodic, bactericidal effects [17]; [11] and antifungal effects [10]. More so, saponins have been found to intercalate DNA of microorganism as mechanism of their antimicrobial activity [7] and thus exhibit antifungal activity [2].

The phytochemical test of the second plant parts, the leaf extracts of *Cleistopholis patens Benth* also revealed the presence of glycosides, steroids, terpenoids, alkaloids, saponins, flavonoids and carbohydrates. Acidic compound and proteins were not detected in all the extracts. The methanol leaf extract of this plant showed the presence of copious amount of terpenoids and saponins, with substantial amount of glycosides, steroids, alkaloids and saponins in the same extract. Hot water and n-hexane extracts of this plant parts revealed the absence of most important secondary metabolites known to have antimicrobial activity [3], [2], [17] and [11]. Even where these important metabolites occur, they exist at low level. The absence of major metabolites in the hot water and n-hexane leaf extracts of *Cleistopholis patens Bent* might be the reason for their negligible antimicrobial activities against the test microorganisms.

Futher evaluation, isolation and characterization of these metabolites are therefore encouraged in order to elucidate the active agents responsible for the observed numerous medicinal effects of these plants.

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