

Short Communication

PHYTOCHEMICAL ANALYSIS OF N-HEXANE LEAF EXTRACT OF *ALPINIA PURPURATA* (VIEILL.) K. SCHUM USING UV-VIS, FTIR AND GC-MS

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ABSTRACT

**Objective:** The present study was carried out to characterize bioactive constituents present in n-hexane leaf extract of *Alpinia purpurata* (Vieill.) K. Schum.

**Methods:** Phytochemical screening of the leaf extract of *Alpinia purpurata* revealed the presence of some bio-active components. The crude extracts were scanned in the wavelength ranging from 200-800 nm by using Ultraviolet-Visible (UV-Vis) spectrophotometers. Fourier transform infrared spectrophotometer (FTIR) was used to determine the functional groups in the plant. Gas chromatography-mass spectrometry (GC-MS) analysis was also performed to find major phytoconstituents present.

**Results:** The phytochemical tests showed the presence of alkaloids, terpenoids, flavonoids, steroids, cardioglycosides, oils and fats, tannins and carbohydrates in n-hexane leaf extract of *A. purpurata*. In UV-Vis analysis there were sharp peaks from 200-700 nm. In FTIR analysis, the plant showed the presence of ester carbonyl and unsaturated carbonyl groups in 1708 and 1691  $\text{cm}^{-1}$  respectively. There were strong absorption bands at 2927 and 1452  $\text{cm}^{-1}$  due to CH and  $\text{CH}_2$  groups. The GC-MS analysis revealed the presence of different phytochemical compounds. This is the first time the presence of 4-Morpholinomethyl-7-methoxycoumarin 1.42%, Methanesulfonate of (3R,4S)-3-Propargyloxy-4-[(R)-1-hydroxy-3-phenyl-3-butenyl]-1-(p-methoxyphenyl)-2-azetidinone 2.28%, 5-Butyl-3-Methyl-1,2,3, 8a-Tetrahydroindolizine 6.48%, Phenol, 4-(3,7-dimethyl-3-ethenylocta-1,6-dienyl)-6.99%, 1-Naphthalenepropanol,  $\alpha$ -ethenyldeca hydro- $\alpha$ ,5,5,8a-tetramethyl-2-methylene-[1S-[1 $\alpha$  (R\*),4 $\alpha\alpha$ , 8 $\alpha\alpha$ ]]-9.29%, Methenolone 10.93%, and Nonanamide, 5-hydroxy-5-methyl-2-(2-methylpropyl)-N-benzyl-25.80% were reported on the leaf extract of *Alpinia purpurata*.

**Conclusion:** From the results, it is evident that *A. purpurata* has various phytoconstituents and functional groups. The intensive study of the resultant active constituents will lead to the discovery of a novel botanical-drug.

**Keywords:** *Alpinia purpurata*, N-hexane, Phytochemical screening, UV-VIS, FTIR, GC-MS analysis.

Zingiberaceae is one of the largest family in the plant kingdom. It is an important natural resource that provides many useful products for food, spices, medicines, dyes, perfume and aesthetics to man. *Alpinia* is the largest genus of the family with more than 200 species [1], with vast majority of them possessing important biological activities. *Alpinia purpurata* (Vieill.) K. Schum (red ginger) is a herbaceous perennial plant, internationally known in the ornamental plant market as potted plant, landscape accent and cut flower [2, 3]. The rhizome has sharp odour, which could improve appetite, taste and voice. It is also used for headache, rheumatism, sore throat and renal disease [4]. The MABA (Microplate Alamar Blue Assay) assay results of the crude ethanolic extract of the various parts of *A. purpurata* have shown the leaf extracts to possess the highest activity, followed by the rhizome and flower extracts [5]. The plant possesses moderate antibacterial and anticancer activities, which may be due to the presence of secondary metabolites in the leaves of *A. purpurata* [6]. In addition to the proposed anti-inflammatory activity, its phytomedicinal potential to treat tuberculosis is also described [5]. In the present work, FTIR and GC-MS technique was used to identify the functional groups and to investigate the phytoconstituents present in the n-hexane leaf extract of *Alpinia purpurata* respectively.

The leaves of the plant *A. purpurata* were collected from the natural habitats of Kanyakumari district, Tamil Nadu, India. The plant specimen was authenticated by Dr. G. V. S Murthy, Botanical Survey of India, Coimbatore, TNAU Campus, India. The leaves were washed thoroughly in tap water, shade dried and powdered. The powder (100g) was exhaustively extracted with n-hexane in the ratio of 1:5 (w/v) for 24 h by using soxhlet apparatus. The extract was evaporated to dryness using the rotary flash evaporator (Buchi type).

Preliminary phytochemical screening of n-hexane crude extract of *A. purpurata* was estimated according to the method adopted by Paech

et al. [7]. The leaf extract showed the presence of most of the secondary metabolites from the solvent, as summarized in [table 1].

Table 1: Phytochemical screening of n-hexane leaf extract of *A. purpurata*

Extract	AL	SA	TP	FL	ST	CG	OF	TN	AP	CH
n-hexane	+	-	+	+	+	+	+	+	-	+

“+” means present; “-” means absent.

AL: Alkaloids; SA: Saponins; TP: Terpenoids; FL: Flavonoids; ST: Steroids; CG: Cardioglycosides; OF: oils and fats; TN: Tannins; AP: Amino acids and Proteins; CH: Carbohydrates

Sample (100 $\mu$ l) was made up to 3 ml by adding n-hexane and then scanned in the range of 200 to 800 nm by using UV-Vis spectrophotometer (Model-Shimadzu UV2450). The spectrum peaks are at 284 and 670 nm with the absorption of 0.588 and 0.002 respectively [fig. 1(a)].

The n-hexane extract of *A. purpurata* was mixed with KBr salt, using a mortar and pestle, and compressed into a thin pellet. Infrared spectra were recorded on a Shimadzu FTIR spectrophotometer 8000 series, between 4,000-400  $\text{cm}^{-1}$ . The FTIR spectrum profile of *Alpinia purpurata* is illustrated in the [fig. 1(b)]. The C=O absorption bands at 1708 and 1691  $\text{cm}^{-1}$  show the presence of ester carbonyl and unsaturated carbonyl groups respectively. The strong absorption bands at 2927 and 1452  $\text{cm}^{-1}$  are due to CH and  $\text{CH}_2$  groups respectively [8]. The spectrum confirmed the presence of alcohols, phenols, alkanes, alkyl halides, carboxylic acids, aromatics, nitro compounds and amines in the n-hexane extract.

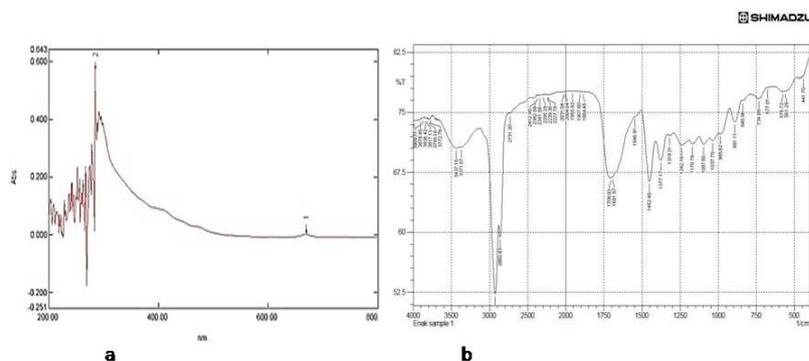


Fig. 1: (a)-UV-VIS spectrum and (b)-FTIR Spectrum of n-hexane leaf extract of *Alpinia purpurata*

The phytochemical investigation of n-hexane extract of *Alpinia purpurata* was performed on a GC-MS equipment (Thermo Scientific Co.) Thermo GC-TRACE ultra ver: 5.0, Thermo MS DSQ II. Experimental conditions of GC-MS system were as follows: DB 5-MS Capillary standard non-polar column, dimension: 30 Mts, ID: 0.25 mm, Film thickness: 0.25  $\mu$ m. Flow rate of mobile phase (carrier gas: He) was at the rate 1.0 ml/min. In the GC part, temperature programme was 70  $^{\circ}$ C raised to 260  $^{\circ}$ C at 6  $^{\circ}$ C/min and injection volume was 1  $\mu$ l. Samples dissolved in chloroform were run fully at a range of 50-60 m/z and the results were compared by using Wiley Spectral library search programme.

The GC-MS spectrum confirmed the presence of various components with different retention times as illustrated in [fig. 2]. The mass spectrometer analyzes the compounds eluted at different times to identify the nature and structure of the compounds. The large compound fragments into small compounds giving rise to appearance of peaks at different m/z ratios. These mass spectra are finger print of that compound which can be identified from the data library [9].

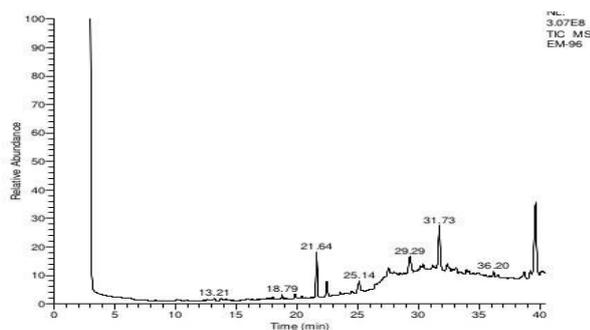


Fig. 2: GC-MS Chromatogram of n-hexane leaf extract of *Alpinia purpurata*

The results pertaining to GC-MS analysis of the n-hexane extract of *Alpinia purpurata* lead to the identification of a number of compounds. The various components present in the leaf extract are as follows 11,23-Di-tert-butyl-5,17-diethoxycarbonyl-25,26,27,28-tetrahydroxycalix[4]arene, Phenanthrene, 3-methyl-(CAS), trans-13-Octadecenoic acid, 2,4a,8,8-Tetramethyldecahydrocyclopropa [d]naphthalene, (-)- $\delta$ -Panasinin, Pregn-4-ene-1,20-dione, 12-hydroxy-16,17-dimethyl-, Caryophyllene oxide, Benzene, 1-methoxy-4-(phenyl ethynyl)-(CAS), 8,12-Diethyl-1,9-dioxo-2,3,7,13,17,18-hexamethyl-1,19,22,24-tetrahydro-21H-bilin, 2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-(CAS), Cyclohexane-1-methanol, 3,3-dimethyl-2-(3-methyl-1,3-butadienyl)-, Longi folen aldehyde, 1,4-Cis-1,7-Trans-Acorenone, 2-Penta decanone, 6,10,14-trimethyl-(CAS), 2-hexyl-1-decen-3-yne, (E,E)-Farnesyl acetone, 9,9-Epoxy-methano-6,6-dimethyl-3,4-undecadien-2,10-dione, Squalene, 2,6,10,14,18,22-Tetracosane hexaene, 2,6,10,15,19,23-hexamethyl-(CAS), 2,6,10,14,18,22-Tetracosane hexaene, 2,6,10,15,19,23-hexamethyl-(CAS), Synaptogenin B, 2-

Hexadecen-1-ol, 3,7,11,15-tetramethyl-[R-[R\*,R\*-(E)]]-(CAS), Tridecanedial, Sandara copimaradiene, Methyl 6-(7-chloroquinolin-2-yl) pyridine [4,5-b]indole-8-carboxylate, Enoxolone, 4,8,12,16-Tetramethylheptadecan-4-olide, 4-Morpholino methyl-7-methoxycoumarin, 03027205002 flavone 4'-OH,5-OH,7-Di-O-glucoside, Nonacosane (CAS), 1-[2-(2-Hydroxyethyl)phenyl]-2-heptanol, N-[3,5-Dinitropyridin-2-yl]proline, Vitamin K Oxide-(18)O, 1,2-Dimethoxy-5,6-dihydrophenanthridin-6-one-4-carboxylate, dl-(3R\*,3aS\*,7aR\*)-7a-(3,4-Dimethoxyphenyl)-3-methane sulfonyloxy-3a-methoxycarbonyl-2,5-dioxo-1-(3-phenylsulfanylpropyl)-2,3,3a,4,5,6,7,7a-octahydroindole, Methyl 2-ethyl-1,2,3,4,6,7,12,12b-octahydro-4-oxoindolo[2,3-a]quinolizine-2-carboxylate, Nonacosane (CAS), 3,4-bis [5'-(2"-Naphthyl)-2'-methylthiophen-3'-yl]-2,5-dihydrothiophene, 5,7-Diisopropyl-3,3-dimethyl-1-(2',4',6'-tri isopropyl phenyl)-2,3-dihydro-1H-2,1-benzazaphosphole 1-oxide, Methanesulfonate of (3R,4S)-3-Propargyloxy-4-[(R)-1-hydroxy-3-phenyl-3-butenyl]-1-(p-methoxyphenyl)-2-azetidinone, 2-(t-Butyl)-3-(dimethylamino)-4,4,6,6-tetrakis (trifluoromethyl)-1,5,2-dioxaphosphorinane, 3-Di phenylphosphino-4,6-diphenyl-. lamda. (3)-phosphinine. 1-Naphthalenepropanol,  $\delta$ -ethenyl decahydro- $\delta$ ,5,5,8a-tetra methyl-2-methylene-, [1S-[1 $\delta$ (R\*),4a $\delta$ ,8a $\delta$ ]], Phenol, 4-(3,7-dimethyl-3-ethenyl-octa-1,6-dienyl)-, 5-Butyl-3-Methyl-1,2,3,8A-Tetrahydroindolizine, Methenolone, Nonanamide, and 5-hydroxy-5-methyl-2-(2-methylpropyl)-N-benzyl-.

*Alpinia purpurata* (Vieill.) K. Schum, a medicinal plant belonging to family Zingiberaceae, was selected for the present study. The FTIR analysis revealed the presence of ester carbonyl and unsaturated carbonyl, CH and CH<sub>2</sub> functional groups. The presence of various bio-active compounds detected after GC-MS analysis justifies the use of the plant for treatment of various ailments.

The presence of bio-active compounds such as Methanesulfonate of (3R,4S)-3-Propargyloxy-4-[(R)-1-hydroxy-3-phenyl-3-butenyl]-1-(p-methoxy phenyl)-2-azetidinone which is an intermediate in the semi-synthetic synthesis of paclitaxel (commercial chemotherapy) [10] and 4-Morpholinomethyl-7-methoxy coumarin (Anti-cancer activity) [11] may suggest why the plant is used as an antioxidant and an anticancer. Methenolone is used to treat aplastic anemia [12, 13]. However, isolation of individual phytochemical constituents and study of their biological activity will definitely give fruitful results and open a new avenue for discovery of a novel drug.

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#### CONFLICT OF INTERESTS

Declared None

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