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Research Article

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GC-MS Analysis of Phytochemical Compounds in *Cajanus Cajan* Leaf

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ABSTRACT

Cajanus cajan is one of the second most dietary legume crops. The quest in search of new alternatives for raw material for food and pharmaceutical industry suggest the use of natural plant products. The ethanol extracts were prepared by simple method of cold extraction, the extracts were characterized by the Phytochemical analysis, Fourier transform infrared spectroscopy (FTIR) and Gas chromatography mass spectroscopy (GC-MS) to identify some fatty acids and heterocyclic compounds present in the leaves extract. The phytochemical analysis revealed the presence of alkaloids, flavonoids, phenol, saponins, tannins and steroids, the study reveal the chemical constituents of Cajanus cajan leaf that these constituents may be behind their medicinal values in phytomedicine.

Keywords: *Cajanus cajan* leaf; FTIR analysis; GC-MS; Phytochemical analysis

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INTRODUCTION

Presently, agricultural by products are being discarded as waste but they can be put into economic use as new alternative for raw materials for food and pharmaceutical industry. *Cajanus cajan* (Pigeon pea) is a perennial legume from the family Fabaceae is a drought resistant crop important for small scale farmers in semi-arid areas where rainfall is low. Pigeon pea contains high levels of protein and important vitamins and is therefore especially important for people living on subsistence diets. The leaves of pigeon pea have been widely used to kill worms, relieve pain and arrest blood, as a *Chinese medicine* [1]. Chemical constituent investigations have revealed that pigeon pea leaves are rich in flavonoids and stilbenes [2,3]. They also contain saponins, conspicuous amount of tannins and moderate quantities of reducing sugar. It is also an important folk medicine in eastern part of India as fresh juice/ boiled leaves are given orally to nullify the effect of intoxication and as a laxative. The leaf paste is applied in oral ulcers and inflammations. The roots of pigeon pea are also found to possesses genistein and genistin, it also contain pinostrobin, longistylin A and longisytlin C which impact anticancer activity [4-6].There is need to know about pigeon pea plant in view of its anatomical, genetical and morphological value in respect to phytomedicine. Hence, in view of importance of *Cajanus cajan* as ethanomedicinal plant, the present work was aimed to assess the possible antioxidant activities of ethanol leaf extract through phytochemical studies, spectroscopy analysis, GC-MS which revealed that chemical constituents of *cajanus cajan* leaf may be behind their medicinal values in phytomedicine.

EXPERIMENTAL SECTION

Materials and Methods

Fresh leaves of *Cajanus cajan* (Pigeon pea) were collected from Uli in Anambra State of Nigeria. The leaves were authenticated at the Biological Science Department, Anambra State University using a herbarium specimen. The leaves were sun-dried for four days and then ground into powder form to increase its surface area. During the extraction process, 30 grams of *cajanus cajan* powder were measured and soaked in 1000 ml of ethanol for 48 hrs.

The mixture was filtered. The filtrate obtained is a mixture of the plant extract and the ethanol. Distillation process was applied to separate the solvent from the extract by evaporates to dryness. The stock solution of the extract was weighed and stored under refrigeration further study.

Phytochmical Analysis

Quantitative analysis for the presence of saponins, tannins, flavonoids, cyanogenic glycosides, alkaloids, phenols and steroids were carried out using the methods of [7].

FTIR Analysis of the *Cajanus cajan* Leaf Extract

After the extraction process, the extract was collected with sample bottles which comprises of pure extract with little quantity of ethanol. Fourier transform infrared spectrophotometer (SHIMADZU Model IR affinity-1, S/N A213747013651) was used to identify functional groups of the pure extract of *Cajanus cajan* leaf. Variations of the FTIR peak numbers were analyzed so as to identify appropriate functional groups in the extract.

GC-MS Analysis of *Cajanus cajan* Leaf Extract

GC-MS analysis was carried out on a Mass Spectrophotometer Model No QP2010 plus Shimadzu, Japan. The carrier gas used was Helium at a flow rate of 0.5 ml/min. 1 µl sample injection volume was utilized. The inlet temperature was maintained as 250°C. The oven temperature was programmed initially at 80°C for 4 min, then an increase to 240°C. And then programmed to increase to 280°C at a rate of 20°C ending with 5 min. Total run time was 90 mins. The MS transfer line was maintained at a temperature of 200°C. The source temperature was maintained at 180°C. The peaks in the chromatogram were integrated and were compared with the database of spectrum stored in the GC-MS library. The analysis was carried out at National Research Centre for Chemical Technology Zaria, Nigeria.

RESULTS AND DISCUSSION

Table 1: Phytochemical constituents of *Cajanus cajan* leaf

Constituents	Qualitative Analysis	Quantitative Analysis
Alkaloids	+++	565
Cardiac Glycosides	-	11
Flavonoids	++	886
Phenolics	++	47
Saponins	+	51
Steroids	++	283
Tannins	++	578

Note: +++ = highly concentrated, ++ = concentrated, + = in trace; - absent or too little to identified qualitatively

Phytochemicals are essential chemicals found virtually in plants and their different parts and at a different concentration. Table 1 above show the presence of alkaloids in good quantity, alkaloids and their derivatives are used as medicinal agents for their analgesic and bactericidal effects [8]. Cardiac glycosides are important due to their hydrogen cyanic acid poison in the body [7]. Flavonoids are very important they are called the free radical scavengers. They prevent oxidative cell damage, protect against all levels of carcinogenesis and anticancer activity [9]. They reduce the rate of heart disease and are distributed groups of phenolic compounds. Flavonoids in leaves are high and could be behind anti inflammatory, anticancer property of the plant which is in agreement with previous literature [10-12]. Saponins are known for formation of foam in aqueous solution, cholesterol binding properties and haemolytic activity [13]. Steroids are regard as antioxidants *in vitro* and have link with reproduction in human [14]. Tannins were noted for astringency and bitter taste, fasten healing of wounds and inflame muscus membrane [15] (Figure 1). Fourier transform infrared spectrophotometer (SHIMADZU, Model: IR affinity-1; S/N A 2137470136 SI) was used to characterize the plant extract, the analysis on the extract shows the variation of the peaks which was used for the determination of the functional groups of the extracts. Peak position 3577.98 cm^{-1} is for sharp free hydroxyl from alcohols, phenols, carboxylics. 3384.98 cm^{-1} is for stretching strong, broad hydroxyl bond from alcohols phenols or carboxylics. 3145.66 cm^{-1} and 301.42 cm^{-1} are for very broad bonds of acids or carboxylics. Wave number 3327.08 cm^{-1} and 3234.4 cm^{-1} are for medium stretching bond of primary amines. Wave bands 1813.96 cm^{-1}, 1717.6 cm^{-1} and 1231.1 cm^{-1} are for very strong or strong bond of anhydrides, aldehydes, acids or esters. Peak values 1659.56 cm^{-1}, 1566.92 cm^{-1}, 1474.28 cm^{-1} are for variable stretching bond for alkenes and arenes. Wave bands 2257.86 cm^{-1} and 2180.66 cm^{-1} are for sharp variable stretching mode of alkynes or nitriles.

From above discussion *Cajanus cajan* leaf extract contain chemical components such as alcohols, phenols, aldehydes, esters, ethers etc which are primary for essential oils, also having good inhibiting abilities.

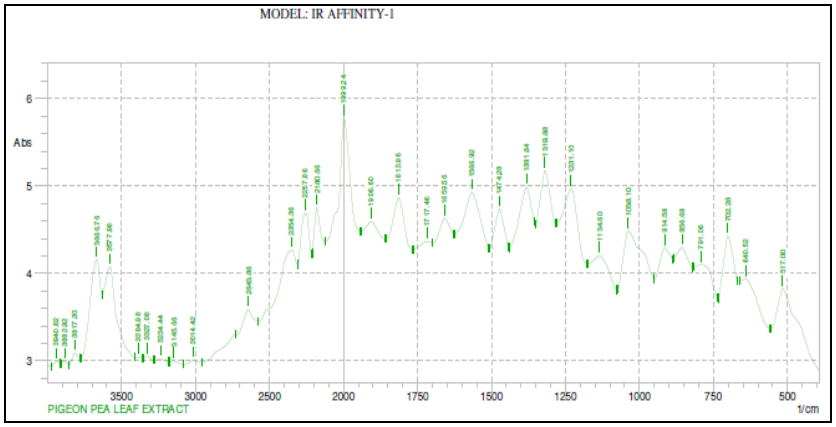

Figure 1: FTIR Analysis of *Cajanus cajan* leaves extract

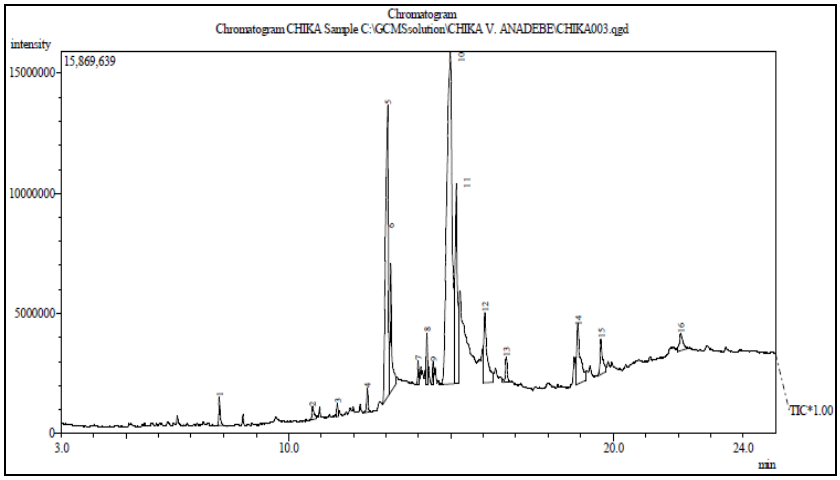

Figure 2: GC- MS Analysis of *Cajanus cajan* leaf extract

Gas Chromatography mass spectroscopy was carried out on the ethanol extract of *Cajanus cajan* leaf .The peaks in the chromatogram were integrated and were compared with the database spectrum of known components stored in GC- MS library. Phytochemical analysis revealed the presence of different fatty acids and heterocyclic compounds. The analysis of the concentrated ethanol extract, resulted many compounds which have diverse use. Compounds having anti-inflammatory, antibacterial, antifungal, skin conditioning properties have been identified (Figure 2).

Peak 1 represent Phenol, 3,5-bis1,1-dimethylethyl, Formula $C_{14}H_{22}O$ Mol Weight 206, Peak 2 indicates Tetradecanoic acid, Formula $C_{14}H_{28}O_2$, Mol Weight 228, Peak 3 represent 1-Octadecyne, Formula $C_{18}H_{34}$, Mol Weight 250, Peak 4 represent Hexadecanoic acid, Formula $C_{18}H_{36}O_2$, Mol Weight 284, Peak 5 indicates n-Hexadecanoic acid, Formula $C_{16}H_{32}O_2$, Mol Weight 256, Peak 6 represent Hexadecanoic acid, Formula $C_{18}H_{36}O_2$, Mol Weight 284,Peak 7 indicates :Oxalic acid, allyl hexadecyl ester, Formula $C_{21}H_{38}O_4$, Mol Weight 354, Peak 8 assigned to 10-Octadecenoic acid, Formula $C_{19}H_{36}O_2$, Mol Weight 296, Peak 9 represent Phytol 2-Hexadecen-1-ol, Formula $C_{20}H_{40}O$, Mol Weight 296, Peak 10 stands for Oxacyclotetradecan-2-one Tridecanoic acid, Formula $C_{13}H_{24}O_2$, Mol Weight 212, Peak 11 assigned to n-Hexadecanoic acid, Formula $C_{16}H_{32}O_2$, Mol Weight 256, Peak 12 represents 1-Tetradecene, Formula $C_{14}H_{28}$ Mol Weight 196, Peak 13 indicates Pentafluoropropionic acid, tridecyl ester, Formula $C_{16}H_{27}F_5O_2$, Mol Weight 346, Peak 14 represent 3,11-Tetradecadien-1-ol Formula $C_{14}H_{26}O$ Mol Weight 210, Peak 15 assigned to 3,11-Tetradecadien-1-ol, Formula $C_{14}H_{26}O$.

CONCLUSION

In this context, phytochemical analysis was conducted on *Cajanus cajan* leaves. These studies revealed the presence of various important bioactive compounds and proved that the plant has commendable sense of purpose and can be

use as a plant of phytopharmaceutical. These results will be of great help in standardization, identification and in carrying out further studies on *Cajanus cajan* leaf based drugs which are used in traditional and modern pharmacopoeia.

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