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Phytochemical evaluation and GC-MS analysis of alkaloid rich fraction of *Millettia aboensis* leaves

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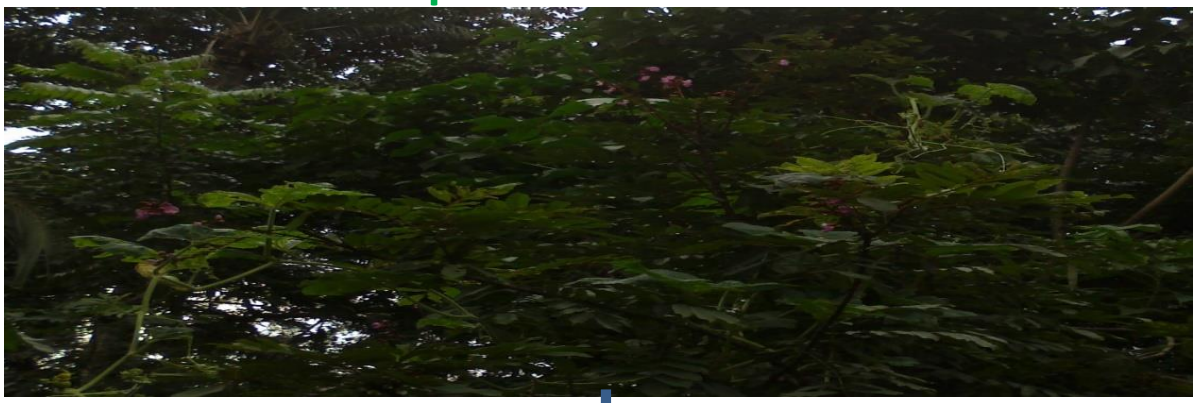
Graphical Abstract



Extraction, Separation and Fractionation

GC-MS

2,4(1H,3H)-pyrimidinedione-1,3,5-trimethyl, 6-cyanoquinoline, pyrimidine-2-amino-6-phenyl, 3-quinolinecarbonitrile, s-Isothiazolecarboxamide 1, 3-dimethyl-4-amino-4,5(1H)-dihydro-1,2,4-triazole-5-one, 3-demethylthiocolchicine

ABSTRACT

Millettia aboensis is a medicinal plant that has been reported to possess several pharmacological properties such as anti-inflammatory, anti-oxidant and anti-microbial activities with no evidence of anti-fertility properties. This present study was designed to investigate the phytocompounds present in alkaloid rich fraction of the of *M. aboensis* leaf in order to ascertain how these bioactive compounds contribute to the anti-implantation and abortifacient activities. The phytochemical result showed abundance of alkaloids, while terpenoids, flavonoids were present in moderate amount. Reducing sugar, steroids and glycosides were sparingly present. The GC-MS analysis of the alkaloid rich fraction revealed the presence of 2,4(1H,3H)-pyrimidinedione 1,3,5-trimethyl, 6-cyanoquinoline, pyrimidine 2-amino-6-phenyl, 3-quinolinecarbonitrile, 3-isothiazolecarboxamide1,3-dimethyl-4-amino-4,5(1H)-dihydro-1,2,4-triazole-5-one, 3-demethyl-3-ethylthiocolchicine. The phytochemical and GC-MS evaluations of alkaloid rich fraction of *M. aboensis* leaf revealed the presence of important bioactive compounds with pharmacological and medicinal properties. Hence, the presence of these phytoconstituents could be responsible for the abortifacient and anti-implantation of the plant.

Key: *Millettia aboensis*, alkaloid rich fraction, Gas chromatography-mass spectrometry analysis, phytochemical, abortifacient and anti-implantation

Introduction

- The traditional folk's use of plants as a source of medicine has been passed down and is now an essential part of the health-care system. Traditional medicine plants include a wide spectrum of chemicals that can be utilized to treat both chronic and infectious disorders(Lakshmi *et al*., 2014).

- *Millettia aboensis* is a member of the Fabacaea family, and among the indigenous people of Nigeria's Nsukka Senatorial District, it is known as "Otoroekpo" or "Uturuekpa." This plant has been claimed to be beneficial in the treatment of constipation, respiratory problems, colds, and headaches (Neuwinger, 2000).

- Tradidionally, this plant has been reported to be a useful plant in the management of constipation, respiratory difficulties, colds and headaches (Neuwinger, 2000). *Millettia aboensis* root maceration in alcohol, on the other hand, has been used to cure hernia and jaundice.

- Gas chromatography mass spectrometry has firmly established itself as a significant technological position for secondary metabolite profiling/studying in both plant and non-plant species in recent years.

- Gas chromatography mass spectrometry has become one of the best techniques in the field of research for identification of constituents of volatile matter, branched chain hydrocarbons, alcohols acids, long chain hydrocarbons, esters and so on (Magashi and Abdulmalik, 2017).
- The goal of this study was to find out what chemicals might be present in the alkaloid-rich fraction of the plant by first making a methanolic extract and then separating and identifying the bioactive compounds using GC-MS.

Results and discussion

Table 1. Phytochemical constituents of alkaloid rich fraction of *Millettia aboensis*

S/N	Phytochemical	Relative abundance
1	Alkaloids	+++
2	Terpenoids	+++
3	Flavonoids	++
4	Steroids	++
5	Reducing sugar	+
6	Glycosides	+
7	Total phenol	ND
8	Tannins	ND
9	Saponins	ND

- Phytochemical screening of alkaloid rich fraction of alkaloid rich fraction of Millettia aboensis leave revealed the presence of the following phytochemicals, alkaloids, terpenoids, flavonoids, steroids, reducing sugar and glycosides

- Some of these phytochemicals identified have been reported to possess antimicrobial, anti-oxidant, anti-fertility, abortifacient, anti-inflammatory, anti-constipation properties and other revealed pharmacological properties.

- Alkaloids have been reported to possess anti-fertility, anti-malarial (Pascal *et al*., 2013)

- Flavonoids have been reported to possess anti-cancer, antiulcer, antispasmodic and other pharmacological properties (Ndukui *et al*., 2014)

Terpenoids also have reported to possess antitumor, anti-inflammatory, antibacterial, antiviral, antimalarial effects, and equally promote transdermal absorption, prevent and treat cardiovascular diseases, and have hypoglycemic activities (Wenqiang *et al*., 2019)

Bioactive Compounds Identified in Alkaloid Rich Fraction of *Millettia aboensis* using GC-MS

S/N	RT (Min)	Name of compound	Molecular formular	M. Wt (g/mol)	Peak (%)
1	38.892	2,4 (1H,3H)-pyrimidinedione,1,3,5 - trimethyl	$C_7H_{10}N_2O_2$	154.17	6.77
2	38.537	6- cyanoquinoline	$C_{10}H_6N_2$	154.17	2.73
3	47.622	Pyrimidine, 2-amino-6- phenyl	$C_{10}H_9N_3$	171.20	67.54
4	7.422	s-isothiazole carboxamide 1,3-dimethyl-4- amino-4,5(1H)-dihydro-1,2,4-triazole-5-one	$C_5H_{10}N_4O$	142.16	2.69
5	34.730	3-quinolinecarbonitrile	$C_{10}H_6N_2$	154.17	34.38
6	35.938	3- demethyl -3-ethylthiocolchicine	$C_{19}H_{21}NO_4S$	359.44	16.15

- According to the article, 2,4 (1H,3H)-pyrimidinedione,1,3,5- trimethyl can be utilized to make primidone, which might cause fetal/neonatal morbidity and mortality during pregnancy. (Drug.com)

- 6-cyanoquinoline has pharmacological properties, as it has been demonstrated to be effective in treating cystic fibrosis caused by the Phe508 mutation with a single medication (Phuan *et al.*, 2011)

- Friedman and Weber-Schöndorfer (2015) found that the pyrimidine analog 5-Fluorouracil (5-FU), which is a pyrimidine analog, interferes with DNA and RNA production.

- However, at least five fetuses or infants have been recorded with significant deformities when the mother was given 5-fluorouracil during the first trimester of pregnancy.

- 3-quinolinecarbonitrile has pharmacological usefulness for single-drug treatment of cystic fibrosis caused by ΔPhe508 mutation (Phuan *et al*., 2011)

- s-isothiazole carboxamide 1,3- dimethyl-4- amino-4,5(1H)-dihydro-1,2,4-triazole-5-one can be used to synthesize Rifapentine, though Rifapentine in pregnant women has not been investigated, but its usage in animal reproduction research have resulted resulted in fetal damage and has been found to be teratogenic. (DailyMed)

- 3- demethyl -3-ethylthiocolchicine Colchicine has been shown to have influence on c-mitotic activity (attenuation of polyploidy) (Evans, 2002).

Conclusion

- This present study has proven that alkaloid rich fraction of *Millettia aboensis* leaves contains some very vital secondary metabolites

- The presence of these phytocompounds could be responsible for the several pharmacological properties such as antioxidant, anti-inflammatory, anti-fertility, abortifacient, antimicrobial, anticarcinogenic, as earlier stated

- The purification of the bioactive compounds and the continued use of the plant in traditional medicine is highly suggested

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