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Ethnobotanical Study and Chemical Analysis of Some Medicinal Plants used in the Management of Breast Cancer in Kebbi State, Nigeria

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ABSTRACT
The use of medicinal plants, especially in Northern Nigeria, plays a vital role in primary healthcare. This paper presents the findings of an ethnobotanical survey on medicinal plants used to treat breast cancer in Kebbi State. Data regarding plant names, the parts used, and preparation methods were collected through oral interviews with herbalists, traditional healers, and rural residents. The information identified 10 plant species from 07 families, with the family Fabaceae being the most prominent, accounting for 30% of the species. The most frequently mentioned plant species included *Prosopis africana*, *Cassia sieberiana*, and *Neocarya macrophylla.* The most commonly used parts were roots, bark, and leaves, with decoctions and concoctions serving as primary preparation methods. Chemical profiling of the three plant extracts was conducted using Fourier Transform Infrared (FTIR) Spectroscopy and GC–MS Analysis. FTIR analysis of the three plants demonstrated the presence of OH acid, C-H (alkane), C=H (alkene), N-H (amine and amide), and C=O (carboxylic acid and amide) groups. Gas Chromatography–Mass Spectrometry identified 29 compounds across the plants. The prominent compounds in *P. africana* are oxalic acid and cyclobutylheptadecyl ester, with 2% of compounds identified. while carbonic acid, tetradecyl vinyl, and 5-Tetradecene (Z)-with 1.97 and 4.94% in *C. sieberiana* and *N. macrophylla*, respectively. The importance of these findings for the search for new botanical options to manage cancer-related issues is discussed.

Keywords: Ethnobotany, Medicinal Plants, Breast Cancer, FTIR GC-MS

INTRODUCTION

For decades, medicinal plants have played a vital role in providing safe and affordable healthcare. In fact, drugs derived from natural products constitute the majority of those approved since 1981 (Zhang *et al.,* 2014). In underdeveloped regions, particularly in Africa, a significant portion of the population still depends on herbal medicine, mainly derived from single plants or plant combinations. This reliance stems from the perceived effectiveness, affordability, and safety of these natural remedies. Ethnobotanical studies on medicinal plants consistently offer opportunities for discovering new compounds from both well-studied and lesser-known plant species. In Nigeria, herbal medicine remains a crucial, cost-effective alternative for treating various ailments, including cancer, microbial infections, and diabetes. Recent ethnobotanical research has identified a diverse range of plants used to treat cancer (Merina *et al*., 2012; Chavan, 2013; Abubakar *et al.,* 2015; Feng *et al*., 2015; Tagne *et al*., 2015; Rumana *et al.,* 2015; Mushtaq, 2017; Abubakar *et al.,* 2020).

Kebbi State, located in Northwestern Nigeria, has a rich history of medicinal plants and a population of over 3.8 million across four emirate kingdoms: Zuru, Argungu, Yauri, and Gwandu. The residents—mainly Dakarkari in Zuru, Gungawa in Yauri, Hausa/Fulani in Gwandu, and Kabawa in Argungu—have relied on traditional plant-based medicines, which have proven effective over time. Although recent studies have highlighted the potential of medicinal plants in Northwestern and Northeastern Nigeria (Kone *et al.,* 2004; Adebayo and Krettli, 2011), there remains a lack of comprehensive research on traditional medicinal practices related to breast cancer treatment in Kebbi State.

Cancer, alongside HIV/AIDS, poses a significant threat to global health, being the second leading cause of death worldwide, responsible for approximately 9.6 million deaths in 2018 (Arnold *et al.,* 2018; WHO, 2020). In 2020, female breast cancer became the leading cause of global cancer incidence, with approximately 2.3 million new cases, representing 11.7% of all cancer cases. It is the fifth leading cause of cancer mortality, resulting in 685,000 deaths. Among women, breast cancer accounts for one in four cancer

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cases and one in six cancer deaths, ranking first in incidence in 159 of 185 countries and first in mortality in 110 countries (Hyuna *et al.,* 2020).
In Nigeria, the cancer mortality rate is notably high; for instance, 51% of breast cancer cases lead to death, compared to 19% in the US. In 2018, Nigeria reported 127,763 cancer cases, with breast cancer being the most common at 39.5%, followed by prostate (14.1%) and cervical cancer (10.7%). Access to cancer treatment in Nigeria is hindered by inadequate facilities and high costs, resulting in many patients never receiving proper care due to delays in diagnosis (Bray *et al.,* 2018; Globacan, 2022). Over 49% of cancer patients resort to complementary and alternative medicine (CAM) (Gaobotse *et al.,* 2023). Conventional treatments such as surgery, radiotherapy, and chemotherapy can have serious side effects, including loss of appetite, nausea, and severe complications like anaemia and infections (National Cancer Institute, 2018). Chemoresistance further complicates treatment (Farley *et al.,* 2015; Atum and Cavalli, 2018), and current synthetic agents often damage healthy cells (Hussein *et al.,* 2014; Ibikunle *et al.,* 2017). There is a need for more research on the efficacy of Nigerian plant extracts as potential anticancer medicines.
Cancer is a global issue and the second leading cause of death in the United States (Kone *et al.,* 2008). In Nigeria, breast and prostate cancer account for 34.2% and 31.7% of deaths among females and males, respectively (Barkaoui *et al*., 2017). The incidence of cancer and cancer-related deaths is continually rising, particularly in underdeveloped regions of the world. This can be attributed to limitations in various treatments, including surgery, chemotherapy, radiotherapy, and others. Additionally, there is a lack of adequate facilities for early detection and the high cost of treatment in these regions. As a result, patients often depend on affordable herbal medicines. Over the years, residents in both rural and urban areas have relied on herbal medicine for cancer treatment, frequently in conjunction with conventional medicine. Patients with mild or advanced-stage cancer tend to withdraw from hospitals to rely entirely on herbal remedies, owing to their potential efficacy, safety, and affordability. Consequently, the present study was undertaken to identify medicinal plants used by herbal medicine practitioners in Kebbi State, thus providing valuable information on herbal practices. Moreover, it is anticipated that the study will uncover new, scarcely investigated plants that could serve as potential sources of novel cytotoxic anti-breast cancer agents. This research aims to conduct an ethnobotanical survey of medicinal plants used for breast cancer management in Kebbi State and to perform chemical profiling of the most frequently cited plant species.

MATERIALS AND METHODS

A cross-sectional ethnobotanical survey was conducted in the study area with authorisation from the Ministry of Environment, through the Department of Forestry, and plants were collected under the approval of the Central Zonal Forestry office. Written Prior Informed Consent (PIC) was acquired, and Access and Benefit Sharing (ABS) was also explained during data collection. Ethnobotanical information was obtained from the traditional healers using a semi-structured questionnaire as described by Dearnley (2005). Some of the questions asked about the origin of knowledge on the plants used in the treatment of breast cancer, the plants' local names, parts used, preparation method, administration, and dosage. In addition, the experience of traditional knowledge outside their community.

The selection of plants for this study was guided by two primary approaches. The first approach was ethnopharmacological, wherein plant samples were chosen based on their recognition for anticancer properties within folkloric medicine. The second approach was chemotaxonomical, which aids in the selection of plant species from genera or families known to yield compounds or classes of compounds exhibiting anticancer activities.

Plant samples were collected with permission from the Zonal Forestry Office, adhering to the rules and regulations governing the collection of medicinal plants as outlined by the World Health Organisation (WHO, 2020). The harvested plant samples were promptly transported to the Herbarium Department of Biological Sciences at Usmanu Danfodiyo University, Sokoto (UDUS) on the same day. Upon arrival, the samples were sorted and cleaned with water to remove any impurities or soil debris. Subsequently, they were cut into small pieces and air-dried under shade on a drying bench for a duration of ten days.

After drying, the plant materials were ground into a fine powder using an electric blender and stored in zip-lock bags in a freezer at −20°C for future analysis. Voucher specimens of the plant species encountered, both in the wild and in home gardens, were prepared. Preliminary identification of the collected specimens occurred on-site, followed by drying according to standard herbarium specimen preparation procedures (Claustro and Madulid, 2005). Initial identification of herbarium specimens was conducted using published reference collections and websites, compared with authentic herbarium specimens

from the Department of Biological Sciences at UDUS. A Botany and Taxonomy instructor ultimately confirmed it.

Fourier Transform Infrared (FTIR) Spectroscopy was conducted in slitless mode using one microliter of diluted samples in methylene chloride, which was injected into the system. The instrument's software employed the "find peak" command to ascertain the frequency and intensity of each spectral band. Each extract was analyzed using a Buck M530 Fourier Transform Infrared Spectrophotometer (FTIR), with the instrument operating over a wavelength range of 4000 to 650 cm^-1.

GasChromatography–Mass Spectrometry Analysis (GC-MS) of the dried ME and Butanol fractions was done by dissolving them in Methanol and Hexane and analysed using a GC-MS model QP2010 plus SHIMADZU with a detector and slit injection system. The temperature was initially set to 60 °C for 3 minutes before progressively increasing to 250 °C. For analysis, 1.6µL of solution was injected at a temperature of 250 °C during the experiment. Helium gas (99.999%) was used as the carrier gas, with a constant flow rate of 1ml/min and an injection volume of 2 µL (split ratio of 10:1); injector temperature 2500 °C, ion-source temperature 2800 °C. The oven temperature was programmed to increase from 1100 °C (isothermal for 2 minutes) at a rate of 10 °C/min to 2000 °C, then 50 °C/min to 2800 °C, and finally to maintain a 9-minute isothermal hold at 2800 °C. Mass spectra were taken at 70 eV, with a scan interval of 0.5 seconds and fragments ranging from 45 to 450 Da. The total GC running time was 36 min. Each component's relative percentage amount was computed by dividing its average peak area by the overall area. The program used to handle mass spectra and chromatograms was Turbo Mass. The identification of components was based on comparison of their mass spectra with those present in the National Institute of Standards and Technology computer data bank (NIST 2009 Library).

RESULTS AND DISCUSSION

This study identifies 10 medicinal plant species from seven families utilised for the treatment of breast cancer in Kebbi State. The Fabaceae family represented the largest share, accounting for 30% of the plants used in cancer management. The survey found that various plant parts are valued in the preparation of remedies for breast cancer, with leaves being the most commonly used part at 50%, followed by stem bark at 30%. The Fabaceae family has been recognised for its anticancer properties (Sebastian *et al.,* 2020), and a similar investigation by Abubakar *et al.* (2020) highlighted *Prosopis africana* as the most frequently cited plant for cancer treatment in Kebbi State. This is consistent with research by Nwafor *et al.* (2024), which also found that leaves are the predominant plant material used for treating various ailments. Additionally, various ethnobotanical studies across other regions of Nigeria (Abubakar *et al.,* 2015; Mainasara *et al.,* 2021) indicated that leaves and stems are the most frequently utilised plant parts in traditional medicine. The preference for leaves and stems may be attributed to their ready availability and the abundance of bioactive compounds, which are known for their medicinal properties. The results, detailed in Table 1, include species names, family names, local names, and the specific parts of the plants used.

Table 1: Medicinal Plants Used for the Treatment of Breast and Uterine Cancer **in** Kebbi State

S/N	Species Name	Family Name	Local Name	Part Use	Preparation and Administration
1.	*Cassia sieberiana.*	Fabaceae	Malga	Leaves	A decoction is taken orally
2.	*Amblygonocarpus Schweinfurthii,* Harms	Fabaceae	Kolo	Leaves	The concoction is taken orally
3.	*Balanites aegyptiaca.*	Balanitaceae	Aduwa	Bark	Powder is applied to cancer, or an infusion is applied to the cells
4.	*Cucurbita pepo,* Dc.	Cucurbitaceae	Kabushi	Fruit	Infusion is taken orally
5.	*Gymnosporia senegalensis*(Lam.) Loes.	Celastraceae	Namijintsada/ kurunkushiya	Root Leaves	Decoction is taken orally or Decoction is taken orally
6. .	*Ziziphus jujuba,* Lam.	Rhamnaceae	Magariya	Seed	Infusion is taken orally
7.	*Neocarya macrophyyla*	Chrysobalanaceae	Gawasa	Leaves	
8.	*Prosopis africana*	Fabaceae	Kirya	Bark	Powder applied topically
9.	*Ximenia americana,* Linn.	Olacaceae	Tsada	Bark	Powder applied as paste topically
10.	*Pistia stratiotes,* Linn	Aroideae	Kainuwa	Leaves	Infusion is taken orally

Additionally, the results of Fourier Transform Infrared (FTIR) Spectroscopy for the most frequently cited species, highlighting the major distinctive peaks, are shown in Table 2. Table 2 presents the primary absorption data from the Fourier Transform Infrared Spectroscopy (FTIR) analysis for the six plants studied. The existence of OH_{acid}, C-H (for alkane), C=H (for alkene), N-H (for amine and amide), and C=O (for carboxylic acid and amide) is demonstrated by the extracts. The functional groups in the extracts suggest that they have similar functional groups and can carry out comparable tasks. Notably, amides are vital in pharmaceuticals (painkillers, antibiotics) and as solvents, due to their stability and diverse properties in nature and medicine. But according to reports, the metal-antibiotic complex can kill germs through a metal-specific mode of action, which is why it is chosen more frequently than organic drugs. The GC-MS of the three most cited plants used in the management of breast cancer **is** presented in Table 3. The result revealed the presence of 29 compounds with 29.35, 11.19 and 5.19% identified in the plant materials. However, multiple RT were observed due to Peak splitting or tailing,the existence of Isomers or Stereoisomers and co-elution of compounds with similar Spectra and duplicates were eliminated using a mechanism in the absence of retention index. Highest peak area, consider the {RT} associated with the highest percentage peak area among all duplicates for the same compound. The peak with the highest area typically represents the true chromatographic maximum and the most concentrated analyte. Where Peak Area percentages are comparable, we consider the {RT} with the highest library match quality. This ensures the final selection is

based on the most statistically confident mass spectral fingerprint, minimising isomer confusion. For any remaining ambiguities or for all major peaks, the final selected {RT} must undergo manual spectral verification against the NIST library and the raw mass spectral data. Crucial for confirming the molecular ion and major fragmentation patterns, validating the selection threshold. The prominent compounds in *Prosopis Africana* are oxalic acid and cyclobutylheptadecyl ester, with 2% of compounds identified. while Carbonic acid, tetradecyl vinyl, and 5-Tetradecene (Z)-with 1.97 and 4.94% in *Cassia sieberiana* and *Neocarya macrophylla*, respectively. Some of these compounds, such as oxalic acid derivatives, have been reported to have antitumor activity in experiments *in vivo* (Fedorov *et al.,* 2018).

Table 2: Resultsof FTIR of the three most cited medicinal plants used in the treatment of breast cancer

A1	3295.0	2922.2	2855.1	1623.3	1032.5
A2	3311.7	2926.0	2847.7	1630.7	1015.7
B1	3291.2	2914.8	2847.7	1623.3	1032.5
B2	3306.1	2918.5	2847.7	1630.7	1032.5
C1	3298.7	2926.0	2851.4	1623.3	1032.5
C2	3311.7	2914.8	2855.1	1623.3	1015.7
Peak range	3300-3400 (dil. soln.)	2850-3000	2850-3000	1630-1695(amides)	1000-1250
Functional groups	N-H (2°-amines)	Csp-H of Alkanes	C-H	C=O (amide I band)	C-N

Key: A1 and A2=Hexane and Methanol extracts of *Prosopis africana***,** B1 and B2 = Hexane and Methanol extracts of *Cassia sieberiana*, C1 and C2=*Neocarya macrophyyla*

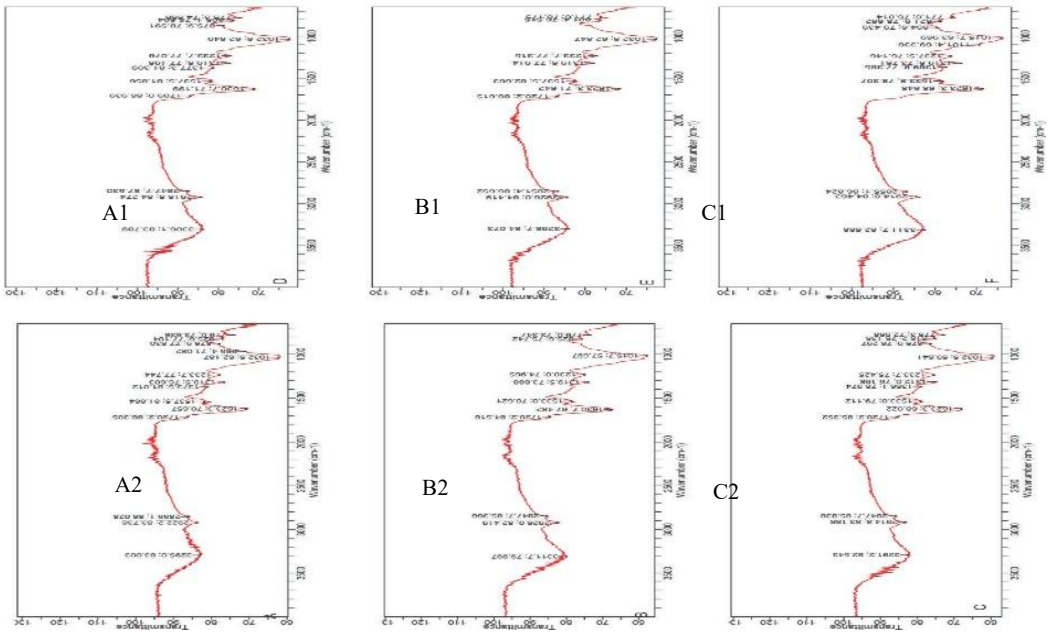


Figure 1:FT-IR spectra of the three plants studied: A1 and A2= Hexane and Methanol extracts of *Prosopis africana***,** B1 and B2 = Hexane and Methanol extracts of *Cassia sieberiana*, C1 and C2 = *Neocarya macrophyyla*

Table 3: GC-MS of the most cited medicinal plants used in the management of breast cancer

S/N	R.T. (Min.)	Compounds	Peak Area % of Samples		
			A	B	C
1	4.102	Carbonic acid, dodecyl prop-1-en-2-yl ester	-	1.06	-
2	4.308	1-Dodecanol, 2-hexyl-	-	0.73	-
3	4.451	1-Tridecene	-	-	0.74
4	4.68	Hexadecane, 1-bromo-	-	1.37	-
5	5.246	Octatetracontane, 1-iodo	-	0.76	-
6	5.67	Nonadecane, 1-chloro-	-	0.86	-
7	6.814	Methoxyacetic acid, tridecyl ester	-	0.71	-
8	6.877	Carbonic acid, eicosyl vinyl ester	-	1.59	-
9	7.106	Cetene	1.07	-	-
10	7.226	Oxalic acid, allyl tridecyl ester	-	1.91	-
11	7.535	7-Tetradecene, (Z)-	-	1.36	-
12	7.81	Tetracontane, 3,5,24-trimethyl-	-	1.53	-
13	8.164	2-Hexyl-1-octanol	1.07	-	-
14	8.239	Carbonic acid, octadecyl prop-1-en-2-yl ester	-	1.82	-
15	10.322	Oxalic acid, allyl undecyl ester	-	0.96	-
16	11.712	Silane, trichlorodocosyl-	-	1.79	-
17	12.444	1-Decanol, 2-hexyl-	1.00	-	-
18	15.637	Oxalic acid, cyclobutylheptadecyl ester	2.07	-	-
19	15.717	Oxalic acid, cyclobutyltetradecyl	0.98	-	-
20	17.589	Methyl 7,9-tridecadienyl ether	-	1.12	-
21	17.663	4-Tetradecene, (E)-	-	-	1.05
22	18.83	Oxalic acid, cyclobutyltridecyl	-	0.81	-
23	19.294	Carbonic acid, tetradecyl vinyl	-	1.97	-
24	22.029	7-Hexadecene, (Z)-	-	-	2.85
25	23.087	Pentacos-1-ene	-	-	1.61
26	23.288	5-Tetradecene, (Z)-	-	-	4.94
		Total %Compounds identified	5.19	20.35	11.19

A= *Prosopis africana* **B=** *Cassia sieberiana. and C=Neocarya macrophyyla*

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