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Research Article

Phytochemicals Screening, Antioxidant and Antimicrobial Capacity of the Leaf Extracts of *Scoparia dulcis*

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Kabir Danjuma*, Kabir Abdu, Imrana Lawan, Moses Jibrin, Magaji Amayindi

Abstract:

Background: *Scoparia dulcis* is a medicinal plant found in low and high temperature areas. **Aims and scope:** This study was done to examine the effect of solvent's polarity on extraction yield, evaluate phytochemicals, antioxidant and antimicrobial capacity of Scoparia dulcis.
Methods: Solvents of different polarities were used to fractionate the ethanol crude by maceration. Phytochemicals screening was done using standard methods, and DPPH experiment was used to screen for antioxidant capacity. Agar diffusion method was used to evaluate the leaf extracts for antimicrobial potential.
Results: Methanol gives highest yield (10.73%), followed by cetone (9.56%), chloroform (6.94%), and petroleum ether (5.16%). Glycosides and anthroquinones were not found, but alkaloids, terpenoids, tannins, saponins, steroids, polyphenols, flavonoids, and reducing sugar were qualitatively detected. *Scopariadulcis* also showed significant antimicrobial capacity; with acetone displaying highest capacity, followed by methanol and chloroform extracts. The results also showed remarkable antimicrobial capacity with acetone exhibiting largest inhibition zone. The extracts also produced Antioxidant capacity with chloroform having highest activity (IC50 = 29.6288 µg/mL), followed by petroleum ether (IC50 = 34.5308 µg/mL), methanol (), and acetone (IC50 = 100.3340 (µg/mL). ANOVA test shows the plant extract has significant antimicrobial and antioxidant capacity at P>0.05.
Conclusion: Scoparia dulcis leaf extracts contain phytochemicals that possess antimicrobial and antioxidant potentials. This research is the first of its kind carried out on this plant in northern Nigeria. The plant is a valuable medicinal plant that can be used for therapeutic applications.

Keywords: Antioxidant Activity, Antimicrobial Activity, Medicinal Plants, Phytochemicals Screening, Scoparia Dulcis.

1. INTRODUCTION

Plant is deemed medicinal if one or more of its organs contains bioactive chemicals that are either therapeutically beneficial or serve as precursors for semi-synthesis of chemotherapeutic medicines. Put otherwise, a plant is considered a medicinal plant if it can be employed as a therapeutic agent, medication, or an active ingredient in pharmaceutical formulations (Shankar et al., 2016).

Medicinal plants, for millennia, have played a key part in African civilization and have been recognized as a symbol of the continent's strong cultural and scientific demands. In order to meet medical and cultural demands while also fostering economic value, the pharmaceutical sector has witnessed a spike in interest in creating herbal treatment procedures, cosmetics made from herbs, and herbal dietary supplements (Eric et al., 2017). Depending on the climate, medicinal plants have been grown and are widely distributed throughout Nigeria. These plants contain bioactive compounds that can be utilized to cure illnesses or as a starting point for the partial synthesis of some helpful medications (Wankupar et al., 2015). In northern Nigeria, medicinal plants have been used by

traditional medicinal practitioners as possible pharmacological agents in the treatment of a variety of illnesses, such as anti-inflammatory, anti-microbial, antioxidant, anti-tumor, anti-diabetic, anti-hypertensive, anti-diarrheal, and anti-cancerous, detoxifying, neuropharmacological, immunity-potentiating agents and other activities (Junaid and Patil, 2020; Haq et al., 2020).

Bioactive, non-nutritive natural substances called phytochemicals are present in the majority of therapeutic plant parts, including fruits, bark, roots, stems, most vegetables, and grains. Such substances may possess biological qualities such as antioxidants and antimicrobials, which could reduce or eliminate the likelihood of oxidative damage from free radicals produced during normal metabolism or microbial assault (Sunayana et al., 2024). Phytochemicals, independently or in combination with other compounds, discover a medicinal plant's therapeutic value. Alkaloids, flavonoids, phenolics, tannins, saponins, steroids, glycosides, and terpenes are important phytochemicals with a range of biological activities found in *Scopariadulcis*. (Junaid and Patil, 2020).

The rapidly growing gap between the public need for new antimicrobial agents and the shrinking capacity for the development of novel therapeutic agents has resulted in a dire situation (Thu et al., 2023). Majority of the achievements in contemporary medicine especially in the treatment of infectious diseases rely on the accessibility of novel infectious drugs and the speed at which this occur is quite very low. The sustainability of therapeutic agents in the treatment of infectious diseases require that new antimicrobial agents are urgently needed as existing therapies lose effect (Ezekwe et al., 2020).

Scoparia dulcis is a perennial herb that grows widely in lower and higher temperature areas. Historically, is gets applications as a remedy for a number of conditions, such as stomach problems, insulin resistant agent, anti-high blood pressure, anti-swelling, pneumonia, hemorrhaging hepatoma, analgesia, antibiotics, antiviral, emmenagogue, antibacterial, antifungal, antiviral, antiherpetic, antiseptic, anti-spasmodic, liver-protective, and anticholinergic agents (Paul et al., 2017). In other ethno-medicine communities in Africa, *Scoparia* herb is also used to treat gastric problems, edema, liver diseases, and respiratory diseases. Other scientific research findings reported anti-cancer activity of *Scopariadulcis*, antidiabetic activity of *Scopariadulcis*, analgesic activity of *Scopariadulcis*, antimicrobial activity of *Scopariadulcis*.

Figure 1. *Scoparia dulcis linn*

Although traditional medicine in northern Nigeria has a long history, it voids of asufficient scientific evidence, particularly in view of current scientific understanding. Antimicrobial and antioxidant agents are still needed, although most pharmaceutical corporations seem to have forgotten about them. This is significant because traditional medicine provides primary healthcare for majority of the world's population (Talal et al., 2019). Scientific literature focusing on the antimicrobial and antioxidant screening of the leaf extracts of scoparia dulcis is scanty, and therefore this study is done to examine how the polarity of the solvents affects the extraction yield, qualitative phytochemical screening, and antioxidant and antimicrobial properties of Scoparia dulcis leaf extracts.

2. MATERIAL AND METHOD

2.1 Sample Collection

The sample material of *Scoparia dulcis* was freshly bought from Zobe Dam area garden, Dutsinma Local Government in the State of Katsina, Nigeria. The Botany Department of Bayero University Kano, Nigeria, verified the authenticity of the plant sample material. A wooden grinder was used to crush the plant sample into powder after it had been cleaned with distilled water and shade-dried. Before being used, the powdered powder was kept in the laboratory (Danjuma et al., 2024).

2.2 Extraction Procedure

Cold maceration extraction was employed. For three days, two hundred grams of powdered *Scopariadulcis* leaf material was soaked in 300 milliliters of petroleum ether solvent while being constantly shaken and stirred. Whatman filter paper was used to filter the mixture. After three days of continuous shaking and stirring, the residue was once more steeped in another 300 mL of chloroform solvent before being filtered again. The procedure was repeated with acetone and methanol solvents, respectively. After the filtrates were concentrated and the residue was eventually disposed of, the dried extracts were measured, and the following formula was used to determine each extract's % yield:

Yield (%) = Weight after evaporation X 100% (1)
Weight of powdered sample

2.3 Phytochemicals Analysis

Phytochemicals (Alkaloids, flavonoids, terpenoids, polyphenols, tannins, saponins, reducing sugar, cardiac glycosides, steroids, and anthraquinones) were all screened from the plant using the usual techniques described below:

Test for Alkaloids (Haager's Test)

2 mL of each extract was added to 2 mL Hager's reagents. The emergence of a milky/creamy white precipitate indicated the presence of alkaloids (Silva et al., 2017).

Flavonoid Test (Alkaline Reagent Test)

Two milliliters of 2% sodium hydroxide solution were mixed with one milliliter of each extract, and then some drops of HCl were added. Flavonoids were indicated by a bright yellow hue that turned colorless when diluted acid was added (Gul et al., 2017).

Polyphenol detection using the iodine test

One milliliter of each extract was combined with a couple of drops of iodine solution that has been diluted. A structure that momentarily became crimson suggested the presence of phenolic compounds (Singh and Kumar, 2017).

Tannins detection using Gelatin Test

Two milliliters of extract and five milliliters of distilled water were mixed with 1 % gelatin solution and 10 % sodium chloride, and the result was a white precipitate that showed the presence of tannins.

Detection of Saponins (Froth Test)

To assess the appearance of saponins, a half mL of each extract was mixed with one mL of distilled water and shaken violently. The production of foam demonstrated the presence of saponins (Junaid and Patil, 2020).

Testing of Steroids (Salkowski's Test)

When two mL of concentrated H_2SO_4 and two mL of chloroform were added to two mL of each fraction, the presence of steroids was indicated by the appearance of yellowish-green fluorescence (Junaid and Patil, 2020).

Testing of Anthraquinones

1 mL of the sample was combined with 10 mL benzene and shaken. The mixture was filtered and 5 mL 10 % (v/v) ammonia was added and shaken again. No visible reaction was observed which confirmed the absence of anthraquinones (Muhammad et al., 2018).

The Benedict Test for Reducing Sugars

0.5 mL of each extract was added to 0.5 mL *Benedict's reagent* and heated by boiling for 2 minutes. A green color indicates the presence of reducing sugar (Singh and Kumar, 2017).

Test for Cardiac Glycosides

In a test tube, 1.5 mL of glacial acetic acid was mixed with 1 mL of the extract. Concentrated sulfuric acid and one drop of 5% ferric chloride were applied to the test tube's side. Since there was no blue-colored solution in the acetic acid layer, cardiac glycoside was not present (Danjuma et al., 2024).

Detection of Terpenoids

A quantity, 5 mL of each sample was combined with 2 mL of chloroform solvent. Three milliliters of H_2SO_4 concentrate were added after the mixture had evaporated on a water bath. The presence of terpenoids was revealed by a grey-colored solution (Gul et al., 2017).

2.4 Antimicrobial Assay

Preparation of the Media

The manufacturer's instructions were followed in preparing the media. To prepare the media, 28 g of Nutrient Agar (NA) and 39 g of Potato Dextrose Agar (PDA) were weighed using a digital weighing balance. The media were then spread out in separate 1500 ml conical flasks, each of which held 1000 ml of distilled water. The conical flasks were promptly sealed with cotton wool, covered with aluminum foil, and heated to complete dissolution. After being autoclaved for 15 minutes at 121^0C, the dissolved media were chilled for 45 minutes at room temperature and then transferred into sterile petri dishes, where they solidified for 24 hours prior to sample inoculation (Kebede et al., 2021).

Collection of pathogenic organisms

Acquired from the microbiology section of the Aminu Kano Teaching Hospital, the test organisms were two bacteria (*Staphylococcusaureus* and *Escherichiacoli*) and two fungi (*Aspergillusflavus* and *Aspergillusniger*). They were then kept in the department of microbiology, Bayero University in Kano, Nigeria

Developing a uniform inoculum

By emulsifying a loop full of colony into a test tube filled with regular saline solution, a suspension of the test bacterium was created. The bacterial suspension's inoculum density was set at 0.5 ml McFarland standard (Kebede et al., 2021).

Preparation of the Stock Solution

Sixteen milligrams of each sample extract were combined with two mL of dimethyl sulfoxide to create a stock solution that contained 8 mg/ml. The conventional dilution formula (C1V1 = C2V2) was used to create concentrations of the fractions at 4000 µg/ml, 2000 µg/ml, 1000 µg/ml, and 500 µg/ml. The standards for

bacteria and fungi were gentamycin (20 µg/ml) and nystatin (50 µg/ml), respectively. A solvent of dilution (DMSO, or dimethyl sulfoxide) served as the negative control (Kebede et al., 2021)..

Bioassay Procedure

The media were cooled at 40 to 45^0 degrees Celsius after sterilization, and then they were transferred into petri dishes. The fractions to be used and the name of the culture were written on the petri plates. The normal bacterial and fungal suspension was dipped into a disinfected cotton-wool swabs (sample stick). Five wells, each 6 mm in diameter, were made in the center of the agar mixture in each petri dish using a sterile Cork borer. The wells were then filled with the prepared extracts and incubated at 37ºC for twenty-four hours. Using a ruler, the zone of inhibition was measured and reported in millimeters (Kebede et al., 2021).

2.5 DPPH Radical Scavenging Assay

The DPPH radical scavenging assay was used in this research to measure *Scopariadulcis's* capacity to scavenge free radicals. 39.4 grams of DPPH were dissolved in 100 milliliters of methanol to create a solution containing 0.1 milligrams of DPPH. In order to make a 1000 µg/ml stock solution, 5 mg of the extract will be dissolved in 5 ml of ethanol. The concentrations 1,000 µg/ml, 500.00 µg/ml, 250.00 µg/ml, 125 µg/ml, 62.50 µg/ml, 31.25 µg/ml, 15.625 µg/ml, and 7.8 µg/ml were created from the stock solution using two-fold serial dilution. The sample and DPPH were incubated for 30 minutes in a 96-well plate. The absorbance at 517 nm was measured using a UV-VIS Shimadzu Corporation spectrometer. The testing was conducted in threefold using the vitamin C as the reference standard. A methyl alcohol and 1-diphenyl-1-picryl hydrazyl mixture bereft of the sample served as a negative control. Using IC50 software (IC50.kt), the IC50 values of the samples and the standard were determined from their percentage inhibitions (Siddartha et al., 2022). The percent DPPH scavenging effect was calculated by the use of the formula below:

$$\text{Inhibition (\%)} = \frac{A_0 - A_1}{A_0} \; 100\ \% \qquad (2)$$

Where A0 is the absorbance of negative control, and A1 is the absorbance of the sample.

DPPH testing is a rapid, simple, and cost-effective method for measuring antioxidant capacity that uses free radicals to evaluate a compound's ability to act as a hydrogen provider or a free-radical scavenger. The process of DPPH testing eliminates DPPH as a stabilized free radical. The free radical DPPH and an odd electron combine to provide a noticeable absorbance, or purple color, at 517 nm. For example, DPPH and antioxidant combine to form DPPH-H, which has a decreased absorption with fewer hydrogen compared to the DPPH. As the quantity of absorbed electrons increases, it decolorizes, making it more radical than the DPPH-H. This decrease in capacity is greatly impacted by decolorization. As soon as the DPPH solutions are combined with the hydrogen atom source, the lower state of diphenylpicrylhydrazine is formed, shedding its violet color (Siddartha et al., 2022).

2.6 Statistical Analysis

The statistics used here were shown as mean of standard deviation. One way Analysis of Variance (ANOVA) was used to either accept or reject the null hypothesis using an online Anova calculator. The values $P<0.05$ were considered statistically significant (Shraddha et al., 2025).

3. RESULT AND DISCUSSION

3.1 Result

Effect of Solvent's Polarity on Extraction Yield

Table 1. Percentage yields of the extracts

Extracts	Solvents	Color	Weights (g)	Yield (%)
PE	Petroleum Ether	Dark green	10.32	5.16
CF	Chloroform	Dark green	13.87	6.94
AC	Acetone	Dark green	19.11	9.56
ME	Methanol	Dark green	21.45	10.73

Evaluation of Phytochemicals in the Samples

The result of the preliminary phytochemical screening of the extracts was presented in table 2.

Table 2. Phytochemical screening of the extracts

Phytochemicals	Extracts			
	PE	CF	AC	ME
Alkaloids	+	-	+	+
Terpenoids	+	+	+	+
Flavonoids	+	+	+	-
Steroids	+	+	+	+
Saponins	-	+	+	+
Tannins	+	-	+	+
Polyphenols	+	+	-	-
Glycosides	-	-	-	-
Reducing sugar	+	+	+	+
Anthraquinones	-	-	-	-

Key: + = present, - = absent

Antimicrobial Activity of the Leaf Extracts

Antibacterial Activity

Table 3. Antibacterial activities of *Scoparialeave* fractions (zones of inhibition in millimeters)

		Concentrations (µg/ml)				
Fractions	**Isolates**	**500**	**1000**	**2000**	**4000**	**Control**
P/ether	*E. coli*	7	8	9	10	25
	S. aureus	8	9	10	11	29
CF	*E. coli*	10	11	12	13	25
	S. aureus	10	11	13	15	29
AC	*E. coli*	15	16	18	19	25
	S. aureus	15	16	18	20	29
MeOH	*E. coli*	10	14	15	16	25
	S. aureus					29
		12	14	17	18	

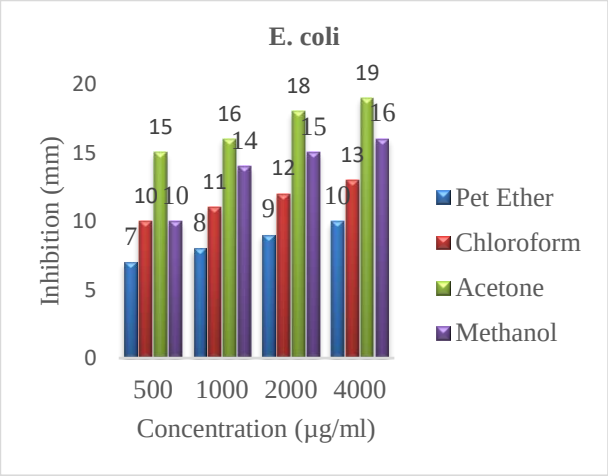

Figure 3. Inhibition zone (*E. coli)*

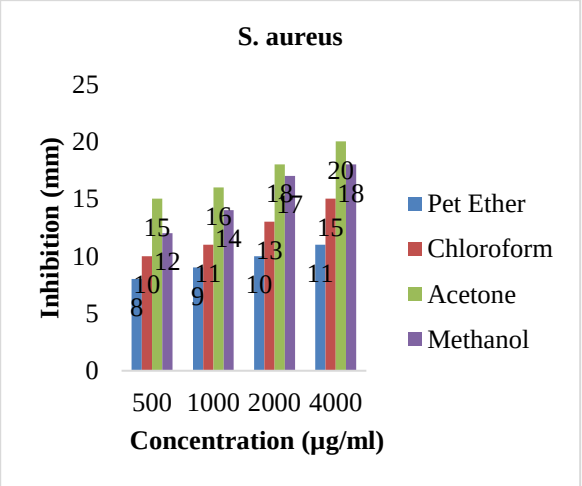

Figure 4. Graph of inhibition for *Staphylococcus aureus* against concentration

Antifungal Property of the Leaf Samples

Table 4. Antifungal Potential of *Scopariadulcis* leaves fractions (zone of inhibition in mm)

		Concentrations (µg/ml)/zones of inhibition (mm)				
Plant' Fractions	**Fungal Isolates**	**50 0**	**1000**	**2000**	**4000**	**Contro l**
Pet ether	*A.flavus*	7	8	9	10	20
	A.niger	8	9	10	11	24
CF	*A.flavus*	9	10	11	12	20
	A.niger	8	9	9	10	24
AC	*A.flavus*	10	12	13	14	20
	A.niger	10	12	12	13	24
MeOH	*A.flavus*	9	10	11	12	20
	A.niger	7	8	9	10	24

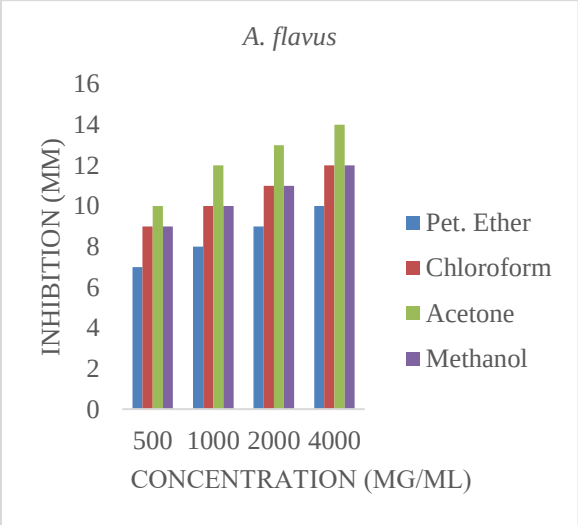

Figure 5. Inhibition zone of *Aspergillus flavus* against concentration

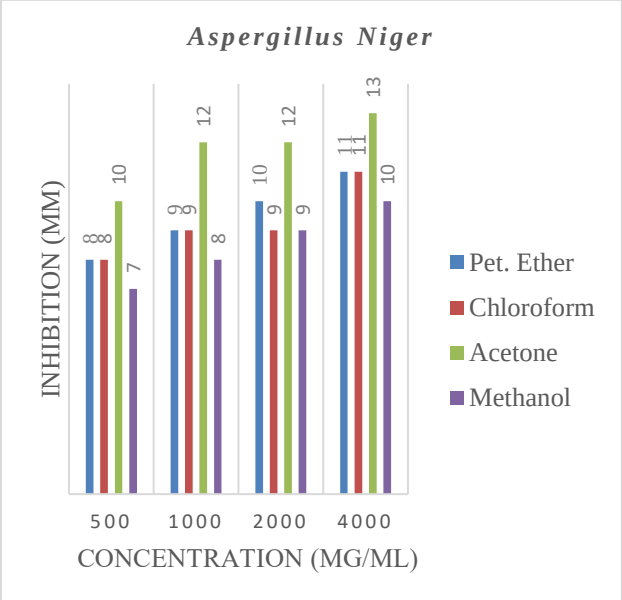

Figure 6. Graph of inhibition against concentration for *Aspergillus niger*

Antioxidant Activity

Figures 7 to 11 present the antioxidant activities of ascorbic acid (standard) and the extracts together with their calculated IC_{50}, respectively.

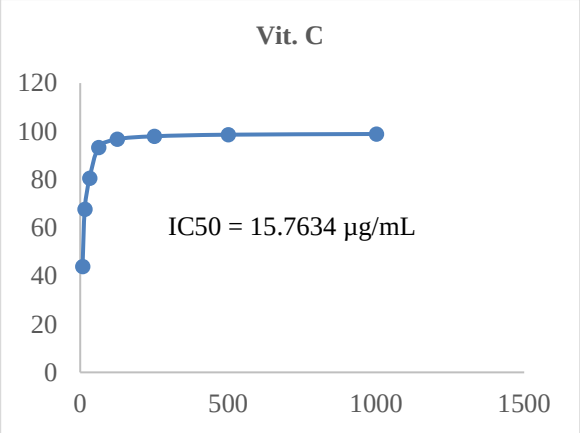

Figure 7. Graph and IC50 of Ascorbic acid and its IC50 (standard)

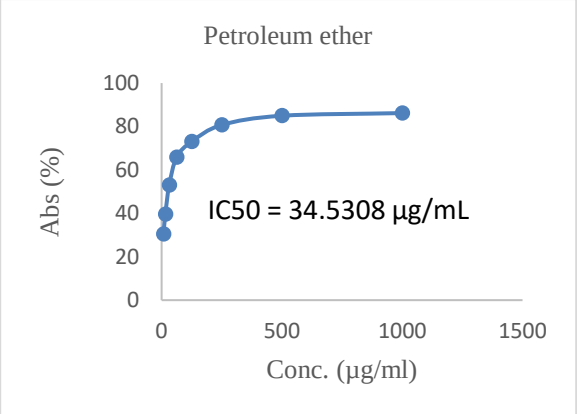

Figure 8. Graph of Absorbance against concentration and IC50 of Petroleum ether

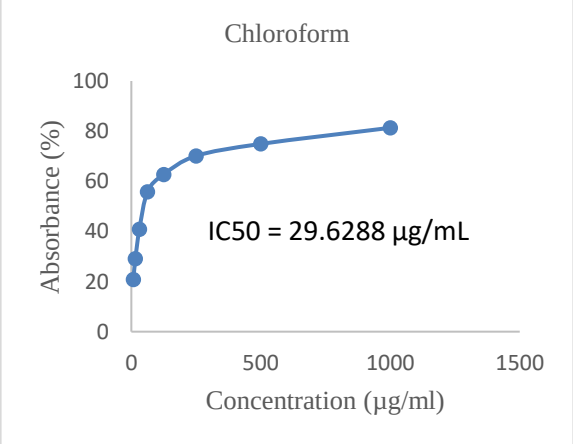

Figure 9. Graph of Absorbance against concentration and IC50 of Chloroform

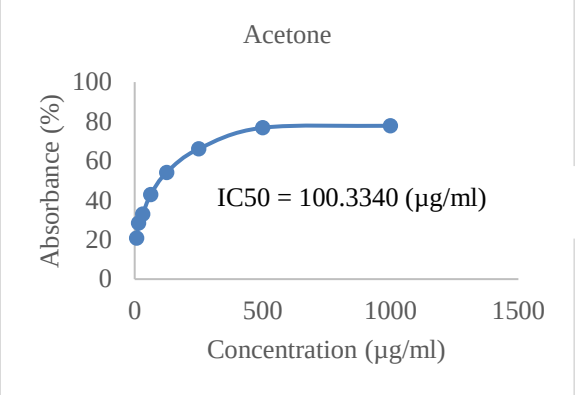

Figure 10. Graph of Absorbance against concentration and IC50 of Acetone fraction

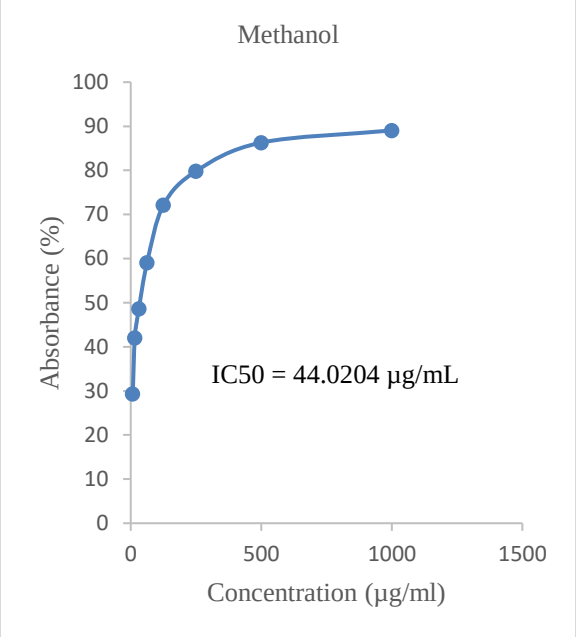

Figure 11. Graph of Absorbance against concentration and IC50 of methanol fraction

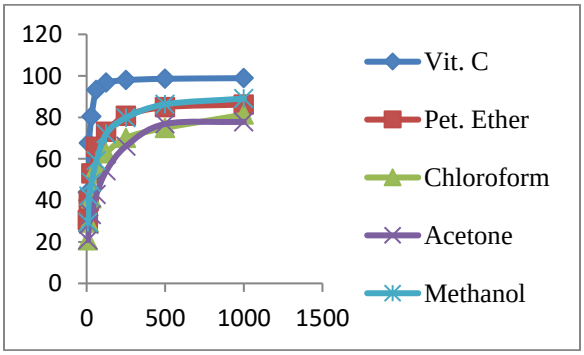

Figure 12. Combined graphs of the extracts

ANOVA Test Analysis

The table 5 below illustrates the Analysis of variance (ANOVA) of the antioxidant activity.

Table 5. Analysis of variance

Test Statistics	Values obtained
Test Statistics F	3.601
P-Value	0.02564

3.2 Discussion

Implications

Polar solvents have the tendency to increase the rate of reactions for polar reactions because of their ability to stabilize charged intermediate and transition state. The extraction of phytochemical constituents and antioxidant compounds from plant material are mostly influenced by factors such as temperature, extraction time, concentration of solvents, and solvent polarity. Phytochemicals are extracted in solvents of different polarities as no single solvent may be trusted to extract all the phytochemicals and antioxidant compounds found in plant (Lesten and Kingsley, 2019).

Research Contribution

The result for the effect of polar, moderately polar, and non-polar solvents on the extraction yields of the leaves extracts of *Scoparia dulcis* was reported in table 1, and it revealed that yield is dependent on polarity. Highly polar solvent (methanol) gives larger yield than moderately polar solvent (acetone). It is evident from these findings that polarity of solvents increases the extraction yields provided that the extraction conditions such as extraction time, temperature, solvent-solute ratio, etc. are kept constant. Recent studies have shown that high polar solvents are highly more likely to extract substantial amounts of phenolics content and antioxidant compounds, thus supporting the idea that polar solvents are more likely to give higher yields of extracts (Shraddha et al., 2025). Another study also reported that solvent polarity tremendously affects the yield of extracts and antioxidant capacity of phenolic compounds in plant (Haq et al., 2020).

Due to its well-known medicinal applications, studies have revealed that several bioactive chemicals have been extracted from various sections of Scoparia dulcis. The phytochemical assessment of *Scopariadulcis* leaf extracts in this study (table 2) shows the presence of reducing sugar, tannins, alkaloids, terpenoids, flavonoids, steroids, and saponins. Anthraquinones and cardiac glycosides were not found in any of the extracts used in this investigation. Studies also found that Scoparia dulcis contains terpenoids, alkaloids, flavonoids, saponins, tannins, and phenols (Muhammad et al., 2018). The presence of amino acids, phenols, saponins, tannins, flavonoids, and terpenoids was demonstrated in other studies (Kebede et al., 2021; Elyaraja et al., 2015). These substances were reported to be bioactive agents in the treatment of diarrhea, fever, hypoglycemia, kidney stones, and stomachaches (Ratnasooriya et al., 2022). These investigations have further validated *Scopariadulcis's* applications in medicine. The presence of tannins, alkaloids, and flavonoids can be the reason for the plant's antimicrobial activity (Aysha et al., 2020).

Table 3 and figures 3 and 4 present the findings for antibacterial properties of the extracts of *Scopariadulcis* in resistance to the tested *E. coli* and *S. aureus* in this study. Theresults show that *Scopariadulcis*leaf fractions contain antibacterial potentials, and it shows that acetone extract possesses a greater antibacterial property in all concentrations; this was followed by methanol extract, chloroform extract, and petroleum ether extracts. It was observed that the inhibition zones increase with increase in concentration. It was also noticed that *S. aureus* shows greater zone of inhibition than the *E. coli*.

Table 4 and figures 5 and 6 reports the antifungal activity of *Scopariadulcis* in this study. The findings show the leaf samples of *scopariadulcis* contains remarkable antifungal capacity against the two pathothenic fungi tested. Acetone extract has stronger antifungal activity (larger zone on inhibition) even at low concentrations than the three extracts. The antifungal activity of the extracts increased with increase in concentrations. *Aspergillusflavus* showed larger zone on inhibitions than the *Aspergilusniger* at lower and higher concentrations.

The antioxidant capacity of the leaf sample extracts of *Scoparia dulcis* has been studied by its ability to reduce DPPH. The outcomes of the DPPH radical testing of the ascorbic acid, and sample extracts of *Scoparia dulcis* in this study were presented in figures 7 to 12. This study revealed that *Scoparia dulcis* leaf has a noteworthy antioxidant and free radical scavenging activity, with ascorbic acid having an IC50 of 9.92.12 µg/ml, followed chloroform (IC50 = 29.6288 µg/ml), Petroleum ether (IC50 = 34.5308 µg/ml), Methanol (IC50 = 44.0204 µg/ml), and then acetone fraction (IC50 = 100.3340 (µg/ml) respectively. A similar research reported that hexane extract of Scoparia dulcis recorded an IC50 of 50.78 ug/ml on both adult male and female Worm.

The Analysis of variance (ANOVA) was used to either accept or reject the null hypothesis, and the results showed the test statisctics F having a value of 3.601 and a P-value of 0.02564 (table 5). This implies that the observed F-test value is statistically significant enough to reject the null hypothesis at an alpha value of 0.05 (5%), and therefore the leaf extracts of scoparia dulcis possess significant antioxidant and antimicrobial capacity.

Limitation

This research is limited to the leaves of Scoparia dulcis. It doesn't account the bioactivity of the stem, barks, or roots of the plant.

Suggestion

We suggest that further similar researches need to be carried out on the other parts of *Scopariadulcis*.

4. CONCLUSION

According to the current investigation, solvent polarity does have an impact on extraction yields as long as the extraction conditions remain consistent. The evaluation of phytochemicals in *scopariadulcis* leaf extract confirmed the presence of alkaloids, flavonoids, terpenoids, saponins, polyphenols, tannins, reducing sugar, and steroids. Cardiac glycosides and anthraquinones were tested and found absent. The leaf extracts of *scopariadulcis* also showed remarkable antimicrobial capacity which increases with increase in concentrations. The leaf fractions of *Scoparia dulcis* showed noticeable antioxidant activities with low IC50 compared with the standard. Therefore, *Scopariadulcis* is a valuable herb that can be utilized for therapeutic applications.

5. AKNOWLEDGEMENT

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AUTHORS CONTRIBUTIONS

KD conceived the research idea, designed the experiment, and wrote most of the paper. KA analyzed and interpreted the data and acted as research advisor. IL assisted in the experiment, performed literature reviews and statistical analysis. MA provided research components such as samples, data, and prepares figures and tables. MJ provided grammatical revisions to manuscript and contributed in analysis data tools. All the authors contributed in the interpretation of the research findings and approved the submission of the manuscript.

AUTHORS INFORMATIONS

Corresponding Author

Kabir Danjuma, Bayero University Kano, Nigeria
Orchid : https://orcid.org/0009-0001-1610-8783
Email : kabirdanjumaa@gmail.com

Author

Kabir Abdu, Bayero University Kano, Nigeria
Orchid : https://orcid.org/0009-0007-9607-1872
Email : kabdu.chm@buk.edu.ng

Imrana Lawan, Yusuf Maitama Sule University, Nigeria
Orchid : https://orcid.org/0000-0002-1853-9114
Email : imranlawanadam@gmail.com

Moses Jibrin, Bayero University Kano, Nigeria
Orchid : https://orcid.org/0009-0009-6985-7399
Email : Mosesjibrin20@gmail.com

Magaji Amayindi, Bayero University Kano, Nigeria
Orchid : https://orcid.org/0009-0002-8216-2697
Email : magajiamayindi@gmail.com

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