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ETHNOMEDICINAL USES AND ANTIOXIDANT AND ANTIMICROBIAL POTENTIAL OF *Spermacoce ocymoides* BURM. F.

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ABSTRACT

Medicinal plants have long been a vital source used by humans for preventive and curative needs. However, there is always an urgent to more document these valuables indigenous knowledge. Hence, the current work aimed to identify the traditional therapeutic virtues of *Spermacoce ocymoides* (Burm. F.) and to evaluate its antioxidant and antimicrobial activities. Ethnobotanical data were collected through an open and semi-structured questions inside three villages in the commune of Dinandougou, Mali. Phytochemical composition was determined by qualitative and colorimetric tests. Antioxidant activity was assessed by DPPH radical and phosphomolybdate methods, and antimicrobial activity by agar diffusion. A total of 40 people were surveyed, with 60% of men and 40% women. These people mainly used the "stem-leaves" combination to treat various illnesses, topped by ringworm (54.93%), acne (29.58%) and infected wounds (9.86%). Recipes made from *S. ocymoides* were mostly prepared by trituration (56%) and administered through skin (97%). The highest levels of polyphenols (3.08±0.16 g EAG/100g), flavonoids (0.72±0.006 g EQ/100g) and tannins (1.11±0.03 g EAG/100g) were recorded with hydroethanolic extracts. These extracts also showed the best antioxidant potential with IC_{50} = 715.55±35.55 µg/mL and the largest inhibition diameters (ID) on *Staphylococcus aureus* (ID = 22.67±0.58 mm) and *Candida albicans* (ID = 17.33±0.58 mm). This study shows that *S. ocymoides* extracts are rich in bioactive compounds and endowed with an appreciable antioxidant and antimicrobial potential.

Keywords: Herbal medicine, *Spermacoce ocymoides*, antioxidant and antimicrobial activity

INTRODUCTION

Since Antiquity, plant resources have always played an essential role in the treatment of pathologies affecting human beings. Therefore, the habitants in rural zones have maintained an ongoing relationship with their environment that led to the development of specific indigenous knowledge regarding the medicinal purposes of plants **[1, 2]**. In Mali, the research on medicinal plants has been dynamic since independence years with the creation of National Institute for Research into Traditional Medicine and Pharmacopoeia (INRMPT), formerly the Department of Traditional Medicine (DMT). This Institute prioritized the valorization of traditional medicine resources through scientific investigations and production of Improved Traditional Medicines (ITMs) **[3]**. Nowadays, in collaboration with others research institutions, works carried out by this department allowed to develop some ITMs used in the management of infectious diseases. Some of these ITMs acquired a Marketing Authorization and are part of the list of Malian essential drugs. For instance, “MALARIAL 5” used against uncomplicated malaria and “DYSENTERAL” used against amoebic dysentery. Marketing Authorization files for other formulated ITMs are in progress, such as “SUMAFURA” and “WOLOTISANE” used against malaria, “SAMANERE” against viral hepatitis, “MITRADERMINE” against dermatoses. These ITMs offer to the population, possibilities of effective, available, accessible, and affordable treatments for the common diseases, especially in rural area communities. Regar -ding these offered opportunities, traditional medicine was classified in African as a valuable cultural heritage and a reservoir of knowledge still widely unexploited or underexploited **[3, 4]**.

Among these underexploited medicinal plant species in Mali, *Spermacoce ocymoides* Burm. F., commonly known as "nguèrè ka da" in the local Bamanankan language of the rural commune of Dinandougou in Mali, is a herbaceous plant belonging to the Rubiaceae family **[5]**. The literature reveals that *S. ocymoides* has many folkloric and ethnomedicinal claims in the treatment of numerous illnesses, especially dermatoses. For example, its leaf juice is applied against ringworm and acne, and the sap is used to treat wounds **[6]**. The various parts of the species are also used to treat hookworm infections **[7]**. Pharmacological studies have shown cytotoxic, antifungal, antibacterial and immunostimulant effects **[8]**, conferring anti-dermatological properties to this species **[9]**. Other work has also revealed that the species contains numerous bioactive principles such as

saponines, alkaloids, phenolic compounds **[9-11]**.

S. ocymoides species grow spontaneously in several areas of West Africa **[5]**, particularly in Mali in the rural commune of Dinandougou. It is highly prized by the local population for various therapeutic purposes. According to our sources of information, its ethnomedicinal and pharmacological properties are not yet documented in Mali. So, it is a vital mission to document the rural ethnomedicinal knowledge for cultural preservation, drug development, and better plant resource management. This work was undertaken to document the ethnomedicinal properties of *S. ocymoides* within the rural population of Dinandougou (Koulikoro Region, Mali) and to validate few of its traditional uses.

MATERIALS AND METHODS

Study area

Ethnobotanical survey was carried out in three villages (Dioni, Fatiambougou and Dianguinébougou) located in the rural commune of Dinandougou, Koulikoro region **(Figure 1)**. This commune is situated inside the Sahelian zone in the southern of Mali.

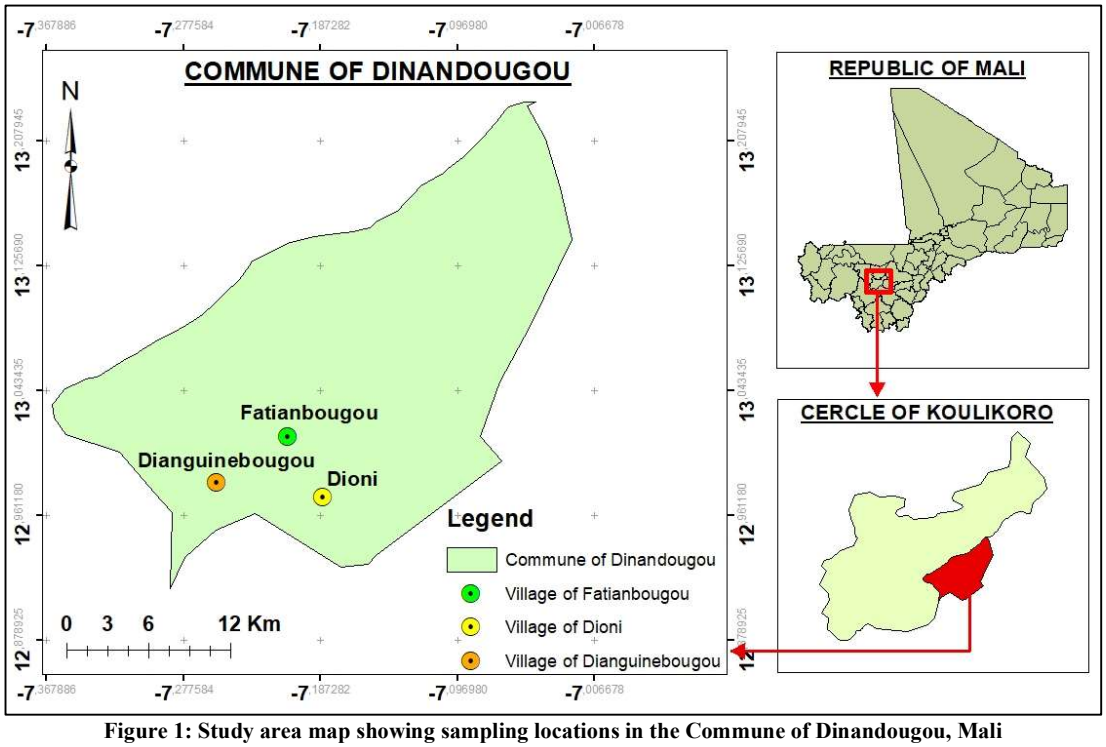

Figure 1: Study area map showing sampling locations in the Commune of Dinandougou, Mali

Plant material

Plant material was consisted of the aerial part (stem and leaves) of *S. ocymoides* harvested at Dioni in Koulikoro in September 2023. After identification and authentication by Dr. Savio Samaké, botanist at the Faculty of Sciences and Techniques (FST), University of Sciences,

Techniques and Technologies of Bamako (USTTB) Mali, the samples were shade dried for three weeks, and then ground into powder samples by using a grinder (Floria®, ZLN3086, 200 Watt).

Biological material

Biological material was consisted of clinical strains of bacteria (*Staphylococcus aureus*) and fungi (*Candida albicans*) provided by the laboratory of "*Clinique Kabala*" located in Bamako.

Ethnobotanical data collection

The survey was carried out based on semi-structured interviews during October 2022 using a survey sheet to identify the therapeutic virtues of *S. ocymoides*. It was conducted with the help of traditional and customary authorities of each village. This accompaniment allowed us to select respondents. Before administering the questionnaire, the aim of the survey was clearly explained to each selected participant in order to obtain their free and informed consent. The informants were designated by the customary and traditional authorities of their village. Interviews were administered in the local language "Bamanankan". The main questions were the illnesses treated by *S. ocymoides*, the parts employed and their modes of preparation and administration. In addition, the sociodemographic characteristics of respondents (age, sex, profession, etc.) were also registered.

Citation frequencies were calculated according to the following formula:

$$\text{Citation frequency (\%)} = \frac{\text{Citation number}}{\text{Total number of citations}} \times 100$$

Ethical considerations

The survey form used in this study was reviewed and approved by the local Ethic Committee of the Biology Department of the Faculty of Sciences and Techniques (FST) of the University of Sciences, Techniques and Technologies of Bamako (USTTB). Interviews were carried out in line with the customs and traditions authorities of each village and with the international standards of the Ethnobiology Society's ethical (ISE Code of Ethics) **[12]**.

Preparation of extracts

Extracts were prepared by maceration in two different solvents (distilled water and 70% ethanol). In an Erlenmeyer flask, 10 g of powdered samples were dissolved in 100 mL of solvent. The mixture was stirred magnetically for 30 min, then filtered under vacuum using wattman paper. This process was operated three times to maximize biocompounds extraction.

Phytochemical investigations

Research of major chemical groups were performed using various qualitative characterization techniques **[13, 14]**. Phytochemical groups were detected by tests:

- ✓ Alkaloids were detected using the Dragendorff/Kraut test
- ✓ Flavonoids by alkaline reagent test

- ✓ Tannins using the Braymer test
- ✓ Coumarins by NaOH test
- ✓ Terpenoids by Salkowski test
- ✓ Anthraquinones by Borntrager test
- ✓ Saponins by the Foam Test

The coloration of each reaction was observed and the presence or absence of targeted phytocompound groups was deduced.

Dosage of total polyphenols

Polyphenol levels were determined by the colorimetric method used by Shatri and Mumbengegwi **[1]**. Briefly, 1 mL of suitably diluted extract or gallic acid solution ranged from 20 to 100 µg/L) was introduced into a flask tube initially containing 800 µL of distilled water. Then 1 mL of Folin-Ciocalteu reagent was added to the mixture. After an incubation of 10 min, a volume of 1 mL of 7% sodium bicarbonate (Na_2CO_3) was added. After homogenization, the solution was immediately diluted with 1 mL of distilled water, and incubated in the dark for 90 min. Absorbances were read against a blank made with distilled water under the same conditions at 750 nm with a spectrophotometer (Thermo Fischer Biomate 3S, Madison, WI53711, USA). Polyphenol contents were expressed in grams gallic acid equivalents per gram of dry weight (g EAG/100 g DM).

Calculations:

The content of polyphenolic compounds was calculated using the following equation:

$$\mathbf{m = \frac{C \times Vf}{Ti} \times Fd}$$

m: Total polyphenol content (mg GAE/g)

C: Concentration of the sample deduced from the standard curve (mg/mL)

Vf: Final volume of the extract (mL)

Fd: Dilution factor

Ti: Test intake (g)

Dosage of flavonoid levels

The quantification of flavonoids was carried out according to the spectrophotometric method reported by Konaré *et al*. **[15]**, which was slightly modified. A volume of 200 µL of extract or catechin solution at different concentrations (20 -100 µg/mL) was introduced into a tube and mixed with 800 µL of distilled water and 50 µL of a 5% sodium nitrite ($NaNO_2$) solution. After 5 min, 50 µL of 10% aluminum chloride ($AlCl_3$) were added, and well homogenized. After 6 min of reaction, 400 µL of 1M sodium hydroxide were added and the reaction mixture was well homogenized. The solution was immediately diluted with 1 mL distilled water and shaken vigorously. Absorbances were read against a blank made under the same conditions with distilled water at 510 nm with a spectrophotometer (Thermo Fischer Biomate 3S, Madison, WI53711, USA). Total flavonoid contents were deduced from a calibration curve previously established with catechin solution using the same equation above. The results were expressed as gram of catechin

equivalents per gram of dry matter (g EC/100 g DM).

Dosage of total tannins

Total tannins were determined using the Folin Ciocalteu method reported by Koutouan *et al*. **[16]**. To 1 mL of plant extract at different concentrations (100 - 500 µg/mL) in a test tube were added 1 mL of distilled water, 200 µL of ethanol, and 100 µL of Folin's reagent, respectively. The reaction solution was homogenized and incubated at room temperature (30 °C) for 5 min. Lastly, 200 µL of 7% ammonium carbonate were added. The solution of gallic acid was used as tannin standard to establish a calibration curve. Absorbances were measured at 725 nm against a blank made of distilled water, after 1 h incubation in the dark at room temperature (30 °C). The concentrations of total tannins were expressed as gram of gallic acid equivalents per gram of dry matter (g EGA/100g) DM.

Assessment of antioxidant potential

The spectrophotometric method was used to perform the antioxidant power with two techniques: DPPH free radical test and phosphomolybdate test.

❖ **DPPH Test**

Free radical 2.2'-diphenyl-1-picryl hydrazyl (DPPH) trapping was used to assess the anti-radical activity of the extracts following the method used by Togola *et al*. **[17]**. A calibration curve was performed with a solution of 100 µg/mL of extract. Fifty (50) µL of each extract at different concentrations were added to 1.95 mL of DPPH methanolic solution (0.024 g/L). A negative control was prepared by mixing 50 µL of absolute methanol with 1.95 mL of DPPH solution. Ascorbic acid, an antioxidant standard, was included in the experiment as positive control. The absorbance was measured using a spectrophotometer (Thermo Fischer Biomate 3S, Madison, WI53711, USA) against a blank prepared for each concentration at 515 nm after 30 min of incubation in darkness and room temperature (30-35°C). The results are expressed firstly in inhibition rates (%), and then translated into IC_{50} (concentration of the tested sample required to reduce 50% of the DPPH free radical) which is inversely proportional to the antiradical activity. The IC_{50} were determined by linear regression from the inhibition percentages as a function of different concentrations of the extracts tested. The free radical inhibition percentages were deduced using the following equation:

$$\textbf{Inhibition rates (\%)} = \frac{\textbf{Absorbance of control} - \textbf{Absorbance of sample}}{\textbf{Absorbance of negative control}} \times 100$$

❖ **Phosphomolybdate test**

The method of Prieto *et al*. **[18]**, recently updated by Konaré *et al*. **[19]** was utilized to assess the total antioxidant capacity (TAC) of our extracts. Extract (100 µL) with 900 µL of working solution (0.6 M sulphuric

acid, 28 mM sodium phosphate and 4 mM ammonium molybdate) were mixed in flask tube. The tube well sealed was incubated at 95°C for 90 min, and then cooled. The absorbance of the solutions as measured at 695 nm using a spectrophotometer (Thermo Fischer Biomate 3S, Madison, WI53711, USA) against a blank containing methanol instead of extract. A calibration curve was generated with ascorbic acid. Lastly, TAC values were deduced from this curve and were expressed as equivalent milligrams of ascorbic acid per gram of dry matter (mg EAA/g DM).

Evaluation of antimicrobial potential

Bacterial and antifungal sensitivity was assessed according to the agar well diffusion technique described by Konaré *et al*. [19], Hawar *et al*. **[20]**.

Preparation of the microbial inoculums

Each microbial strain from the stock cultures was streaked into a petri dish. After 24 h incubation at 37 °C, few colonies from each dish were emulsified in sterile physiological solution of 0.9% NaCl (w/v).

Spectrophotometric method was employed to adjusted the turbidity of each inoculum was then adjusted to 10^8 cells per mL (0.5 McFarland scale). About 2 mL of microbial suspension were read at 620 nm (and properly diluted if applicable) until obtained an optical density of 0.100 around, corresponding to 10^8 CFU/mL on Mac Farland scale.

Disk preparation

A concentration of 4 mg/mL of extract were dissolved into 5% DMSO was used. The blank discs (from Liofilchem SRL) with diameter of 5 mm were filled with with 100 µL of the extract solution (4 mg of extract). Commercial antibiotic discs (doxycyclin at 30 µg/disk and Casprofungin at 5 µg/disk; purchased from Liofilchem, S.r.l., Rogeto degli Abruzzi, Italy) were used as positive control for *S. aureus* and *C. albicans*, respectively. A solution of 5% DMSO (100 µL/disk) was employed as negative control.

Disk diffusion using agar medium

A volume of 150 mL of Mueller–Hilton broth (for *S. aureus*) and Sabouraud Dextrose broth (for *C. albicans*) was poured separately into Petri dishes. Each Petri dish was loaded with 100 µL of microbial suspension and then with discs soaked in plant extract solution. Standard antibiotic disc (doxycyclin and Casprofungin), and blank disc loaded with only the solubilization solvent (without plant extract) were also placed on the top of separate agar Petri dishes for positive control and negative control, respectively. All Petri dishes were firstly kept at 4° C for 1 h to allow the pre-diffusion of extracts through the agar, before being incubated under aerobic conditions at 37°C for 48 h. After the end of incubation, fungal and bacterial growths were observed, and the diameters of inhibition zones (ID) were measured in mm **[19, 20]**.

The appreciation scale reported by Konaré *et al*. **[19]** was used to classify the recorded diameters: if ID < 8 mm, the bacterial strain is considered not sensitive to the extract tested, if 8 ≤ ID < 14 mm, sensitive strain, if 14 ≤ ID < 20 mm, very sensitive strain and if ID ≥ 20 mm, extremely sensitive strain.

Statistical analysis of data

The entry of data and statistical tests were performed with IBM SPSS Statistics version 21 (IBM Corp., Armonk, NY, USA) and Minitab statistical software version 18.1 (Minitab Inc., PA., USA). Qualitative variables were reported in terms of percentages, while quantitative ones were described in terms of means ± standard deviations. The relationship between some variables was searched using the test of Chi^2.

RESULTS

Ethnobotanical data

Sociodemographic characteristics of respondents

The **Table 1** represents the frequency quotes of informant repartition based on the gender, profession and provenance. These educational levels are described in the **Table 2**.

A total of 40 people were questioned, 24 of them were male (60%) and 16 women (40%). Most of the questioned people (60%) were farmers, while 37.50% were doing housework as activity **(Table 1)**. Data from **Table 2** shows that about 60% of the people from the investigated villages were illiterate, i.e. could neither read nor write. Among these illiterates, 35% were men against 25% of female. For respondents who attended school (40%), the fundamental level was predominant (22.50%), followed by secondary one (7.50%). In addition, very few respondents (5%) have benefited from an alphabetization program (informal education).

Use of plant parts and diseases treated by *S. ocymoides*

The citation frequencies of diseases treated, different plant parts used, in addition to their preparation and administration modes are depicted in the **Figures 2, 3,** and **4**, respectively.

Different parts of *S. ocymoides* parts were mainly used to treat dermatological diseases and microbial infections. It was most commonly used to treat ringworm with 54.93% of cases, acne with 29.58% and wounds with 9.86% (**Figure 2**). Most of the selected informants (i.e., 67.50%) used the leafy stem ("stem-leaves" combination), followed by the leaves (i.e., 17.50%) (**Figure 3**). To treat these diseases, most of respondents (56%) prepared their recipes by trituration (**Figure 4A**). Application to the skin (cutaneous route) was the main route of administration (97%) (**Figure 4B**).

Phytochemical composition

The **Table 3** shows the results of phytochemical screening of aqueous and hydroethanolic extracts.

These data indicate valuable insights into the chemical composition of different extract from *S. ocymoides*, exhibiting significant presence of many major groups, such as alkaloids, flavonoids, tannins, coumarins, terpenoids, and anthraquinones. On the other hand, the saponins was not detected in the hydroethanolic extract.

Total polyphenol, flavonoid and tannin contents:

The results for polyphenol, flavonoid and total tannin content expressed for 100 g of dry matter (DM) are given in the **Table 4** below.

Analysis of this table shows that the levels of total polyphenols (p-value = 0.002 < 0.05), flavonoids (p-value = 0.1E-6 < 0.05) and tannins (p-value = 0.0003 < 0.05) varied significantly from one extract to another. Hydroethanolic extracts showed the highest contents of polyphenols (3.08±0.16 g EAG/100g DM), flavonoids (0.72±0.006 g EQ/100g DM) and tannins (1.11±0, 0.03 g EAG/100g DM).

Antioxidant potential

Table 5 summarized the extract ability to scavenge free radical DPPH and the total antioxidant capacity (TAC).

The data from this table reveals that the free radical scavenging capacity of our extracts varied from one extract to another (p-value < 0.05). The hydroethanolic extract exhibited the highest DPPH free radical reducing power (715.55±35.55 µg/mL) and total antioxidant capacity (3.19±0.15 g EAA/100 g).

Antimicrobial potential

The antimicrobial sensitivity expressed in terms of diameter (mm) of inhibition zones (DI) are showed in **Table 6**.

Based on the scale used by Konaré *et al*. **[19]**, all the extracts have been found to be sensitive towards the tested strains a part of *S. aureus* with the aqueous extract. This sensitivity varied according to the microbial strains and to the extract types (p-value < 0.05). While *S. aureus* showed resistance with aqueous extract (06.67±0.58 < 8 mm), it has been found extremely sensitive with hydroethanolic extract recording the highest DI = 22.67±0.58 > 20 mm.

Correlation between antioxidant and antimicrobial activities and phenolic compounds

The previous results revealed the richness in phenolic compounds, natural antioxidant agents and antimicrobial activity. Thus, the correlation plot based on Pearson's coefficient (R^2) was used to detect any correlations between bioactive compounds and biological activities recorded in the extracts. These results are shown in **Table 7**.

There is a very strong correlation between free radical scavenging activity and total polyphenol, flavonoid, and tannin contents, and between these phenolic compounds and antimicrobial activity (R^2 > 0.90). An increase in polyphenol, flavonoid or tannin

levels leads to an increase in DPPH free radical scavenging ability (IC_{50}) and total antioxidant capacity (TAC), with a parallel increase in antimicrobial potential (antibacterial and antifungal). This link between anti-free radical activity and phenolic compounds is largely dependent on flavonoids ($R^2 = 0.99$).

Table 1: Repartition of questioned people per gender, profession and locality (n = 40)

	Sex			Profession			
Sites	Male	Female	TOTAL	Farmer	Breeder	Housewife	TOTAL
Dioni	30%	22.50%	52.50%	32.50%	0%	20%	52.50%
Dianguinébougou	15%	07.50%	12.50%	12.50%	02.50%	07.50%	22.50%
Fatiambougou	15%	10%	25%	15%	0%	10%	25%
TOTAL	60%	40%	100%	60%	02.50%	37.50%	100%

Table 2: Levels of education (n = 40)

Educational levels	Villages of survey			Gender		Total per level
	Dioni	Dianguinébougou	Fatiambougou	Male	Female	
None	35.00	5.00	20.00	35.00	25.00	60.00
Fundamental	7.50	10.00	5.00	12.50	10.00	22.50
Secondary	5.00	2.50	0.00	7.50	0.00	7.50
University	0.00	5.00	0.00	2.50	2.50	5.00
Informal	5.00	0.00	0.00	2.50	2.50	5.00
Total per village/gender	52.50	22.50	25.00	60.00	40.00	100.00

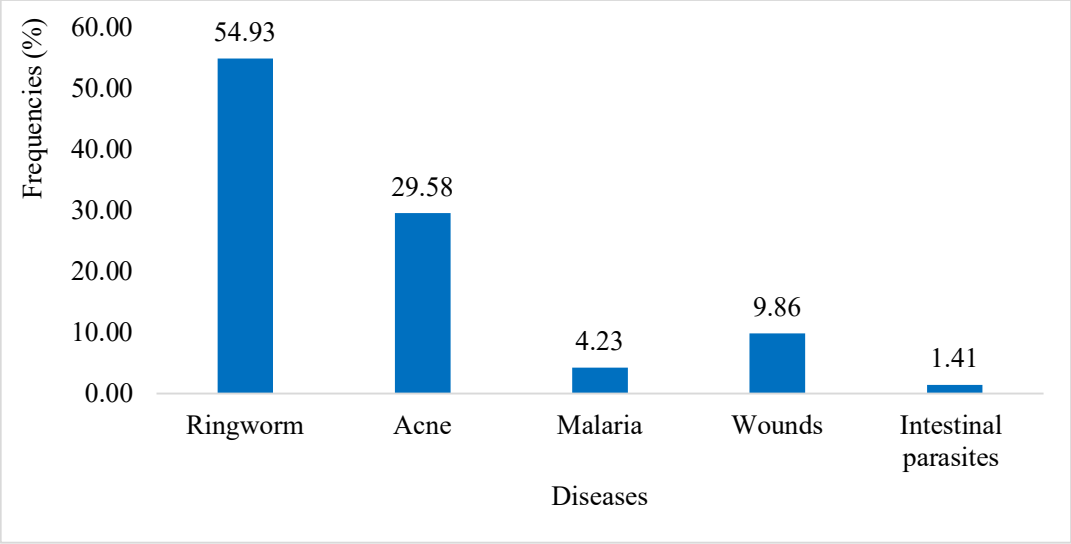

Figure 2: Citation frequencies of diseases treated by *S. ocymoides*

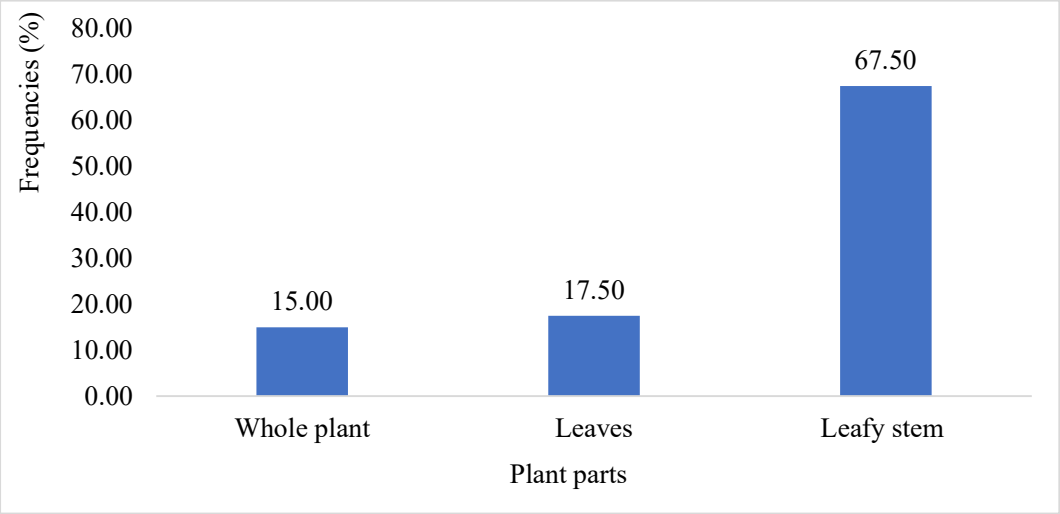

Figure 3: Use frequencies of different parts of *S. ocymoides*

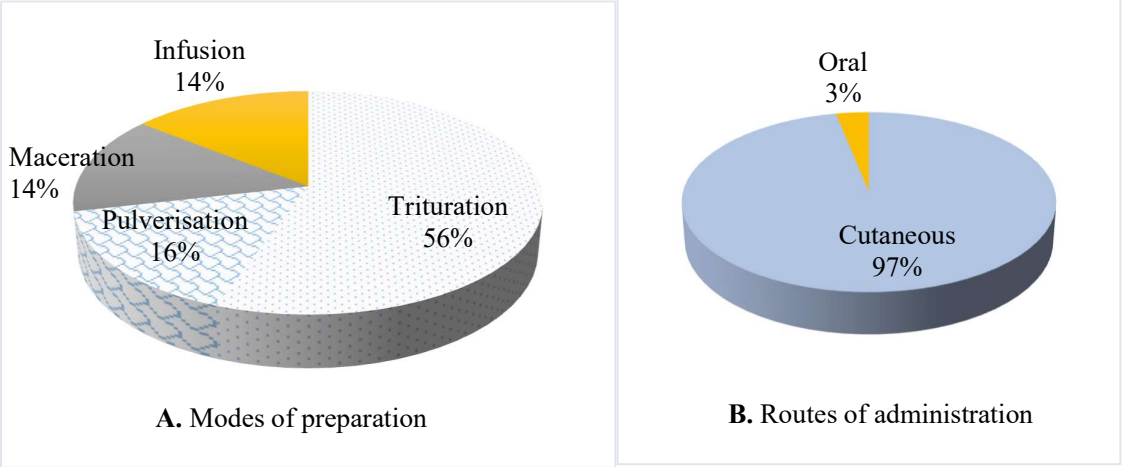

Figure 4: Modes of preparation (A) and administration (A) of recipes made from *S. ocymoides*

Table 3: Phytochemical composition of *S. ocymoides*

Phytoconstituents groups	Extracts	
	Aqueous	Hydroethanolic
Alkaloids	Present	Present
Flavonoids	Present	Present
Tannins	Present	Present
Coumarins	Present	Present
Terpenoids	Present	Present
Anthraquinones	Present	Present
Saponins	Present	Absent

Table 4: Levels of polyphenols, flavonoids et tannins

Extracts	Polyphenols (g EGA/100g) DM	Flavonoids (g EQ/100g) DM	Tannins (g EGA/100g) DM
Aqueous	1.87±0.03^{b}	0.32±0.003^{b}	0.63±0.06^{b}
Hydroethanolic	3.08±0.16^{a}	0.72±0.006^{a}	1.11±0.03^{a}

**For each phytoconstituents, the means that do not share any letter are considered to be significantly different at threshold 0.05*

Table 5: Antioxidant power of extracts

Extracts	IC_{50} (µg/mL)	TAC (g EAA/100 g MS)
Aqueous	3169.19±160.67^{a}	0.29±0.01^{B}
Hydroethanolic	715.55±35.55^{b}	3.19±0.15^{A}

** For each test (DPPH and TAC), the means that do not share any letter are considered to be significantly different at threshold 0.05.*

Table 6: Diameter of inhibition zones (DI) and their appreciation

Extracts and standards	*S. aureus*		*C. albicans*	
	DI (mm)	Appreciation	DI (mm)	Appreciation
Aqueous	06.67±0.58[b]	Non sensitive	13.67±0.58[b]	Sensitive
Hydroethanolic	22.67±0.58[a]	Extremely sensitive	17.33±0.58[a]	Very sensitive
Doxycyclin (Control +)	20	Extremely sensitive	---	
Casprofungin (Control +)	---		18	Very sensitive

** For each microbial strains, the means that do not share any letter are considered to be significantly different at threshold 0.05.*

Table 7: Correlation coefficients of Pearson (R^2)

	DPPH - IC_{50}	Test TAC	Antibacterial potential	Antifungal potential
Polyphenols	0.97	0.96	0.98	0.93
Flavonoids	0.99	0.99	0.99	0.97
Tannins	0.98	0.96	0.96	0.93

**IC_{50}: Inhibiting concentration of 50% of DPPH free radical; TAC: Total antioxidant capacity.*

DISCUSSION

In our survey, the sociodemographic data of the respondents who used *S. ocymoides* for primary healthcare showed that the majority was men and older population. These data fit with a widespread belief in West Africa, as it is the case for all traditional cultures, where older people and men are reputed to be knowledgeable in herbal medicine use **[4, 21]**. As a rule, these men older people have gained precious experience as users of medicinal plants or have learned from other people's experiences **[21]**. The literate rate of the respondents was lower (40%) inside the three villages, while 60% were illiterate. This registered level of education is in line with the national rate of education in Mali, which was 33.7% **[22]**. This slightly high rate of literate could be explained by the presence of an alphabetization program included these three villages in the context of capacity reinforcement of rural populations (informal education). If the level of education was sex-independent (p-value = 0.687 > 0.05), it was influenced by the professional status of respondents (p-value = 0.004 < 0.05). For instance, about 58% of illiterate were farmers.

The respondents used *S. ocymoides* to treat several illnesses leaded by mainly skin disorders, such as ringworm, acne, wounds. In addition, the species use to be employed for malaria treatment, which is an endemic parasitemia in Mali. These data corroborate those from the literature which mentioned the same diseases treated by *S. ocymoides* **[9-11]**.

Nowadays, dermatoses or skin diseases are a major public health problem worldwide. They became one of the main reasons for medical consultation in Mali **[23, 24]**. The hospital prevalence of this pathology rose from 12.75% in 2018 to 15.7% in 2023, mainly affecting the elderly **[24]** and children under five **[23]**. These alarming statistics could justify the use of *S. ocymoides* in the traditional management of skin disorders by the local population of Dinandougou, Mali.

The leafy stem of *S. ocymoides* was the most prized part (67.50%), followed by the leaves (17.50%) to treat these diseases. This strong desire for aerial parts could be explained by the morphological type of *s. ocymoides*, which is a woody herbaceous plant. Importunely, 15% of respondents used the entire plant, which is unsafe for safeguarding the species. Analyses show that this practice is mostly related to educational level, since 67% of illiterates were involved. These results reveal an urgent need to strengthen the educational rate inside. Because, it has been reported that the level of education play an important role in the conservation and survival of valuable natural resources **[25, 26]**.

These parts were mainly prepared by trituration (56%). Few respondents used also granulation (16%), decoction (14%) and infusion (14%). The prepared recipes were mostly administered via cutaneous route (97%). In contrast, the oral route was the least used (3%) by the questioned people. This fact is due to the general form of preparation (trituration) and the main targeted diseases (dermatological disorders). Phytochemical investigation of *S. ocymoides* exhibited the presence of many major groups (alkaloids, flavonoids, tannins, coumarins, terpenoids, and anthraquinones). Interestingly, the hydroethanolic extracts showed the highest contents (p-value < 0.05) of polyphenols with 3.08±0.16 g EAG/100g DM, flavonoids with 0.72±0.006 g EQ/100g DM and tannins with 1.11±0, 0.03 g EAG/100g DM. Through their cytotoxic, antifungal, antibacterial and immunostimulant effects [8, 9, 11], these compounds confer anti-dermatological properties on this species. Many authors' reports on the phytochemical compounds of *S. ocymoides* agrees with our findings on phytochemical profiling **[9-11]**.

Assessment of antioxidant potential revealed that all extracts from this species are endowed with antioxidant power. The hydroethanolic extract exhibited the most interesting ability of reducing free radical DPPH (715.55±35.55 µg/mL) and total antioxidant capacity (3.19±0.15 g EAA/100 g). A high DPPH radical reduction capacity (80%) obtained by Adesegun *et al*. **[27]** with the same species confirms our results. This antioxidant activity of our extracts would be closely linked to phenolic compounds since statistical tests revealed a strong correlation between these phenolic compounds (polyphenols, flavonoids and tannins) and antioxidant activity (R2 > 0.95). Likewise, works of Kähkönen *et al* **[28]** and Catherine *et al*. **[29]** demonstrated that the antioxidant potential of phenols of plant origin was due to the presence of hydroxyl groups on their chemical structure, giving them the ability to scavenge free radicals. These phenolic compounds are also known as 77 effective hydrogen donors, making them good

antioxidants **[29]**. This richness of our extracts in phenolic compounds could make them a potential source of natural antioxidants. As a result, this richness in antioxidant agents could be useful to better neutralize the reactive oxygen species generated in individuals living with skin diseases, since it is well known that phytochemical compounds in a medicine contribute to effectively combat or prevent a disease by their antioxidant power **[30]**.

Skin diseases can be viral, fungal, and bacterial origin. Indeed, it is a bacterial, superficial, epidermal skin infection, non-immunizing, caused by *S. aureus* and *C. albicans* in more than 70% of cases **[9, 31]**. Antimicrobial sensitivity assays showed that all extracts have been found sensitive towards these two strains a part of *S. aureus* with the aqueous extract, based on the scale used by Konaré *et al*. **[19]**. Strain of *S. aureus* was extremely sensitive with hydroethanolic extract recording the highest diameter of inhibition zone (ID = 22.67±0.58 > 20 mm). These ID values are lower than those obtained by Ebana *et al*. **[6]** with the same species, who recorded inhibition zones of 35.0±0.85 mm against *S. aureus*. Similarly, with ethanolic extracts, other studies reported very close results: Oluwayemi *et al*. **[10]** with ID = 10 to 20 mm on *C. albicans*; Minkat *et al*. **[9]** ID = 3 to 15 mm on *S. aureus*. Overall, the best antibacterial and antifungal activities in our study were recorded with hydroethanolic extracts. These results corroborate those in the literature generally attributing the best biological activities of medicinal plants to hydroethanolic extracts **[15, 27, 30]**. The literature attributes this discrepancy in results to the phytochemical composition, influenced by geographical location of the plant as well as sample harvesting stage, and genetic factors **[9, 32]**.

The antimicrobial potential of phenolic and terpene compounds is well documented. The literature reports that the antimicrobial activity of flavonoids is due to their ability to complex with soluble extracellular proteins and the bacterial cell wall, while that of tannins may be linked to their ability to inactivate microbial adhesions, enzymes and cell envelope proteins **[33]**. In addition, our Pearson correlation tests revealed that these compounds were strongly involved (R^2 ranging from 0.93 to 0.99) in the inhibition of bacterial and fungal growth recorded with *S. ocymoides* extracts. This information supports data from of our survey in Dinandougou, Mali revealing the traditional use of *S. ocymoides* in the management of skin disorders mainly caused by the tested microbial strains. Nowadays, the higher rate of antibiotic resistance pattern and their side effects registered toward these pathogens **[1]** calls for an urgent need for discovering new source of effective, affordable and safe molecules.

Furthermore, this work is in accordance with other studies carried out in Africa, reporting that many plants are traditionally used to treat dermatoses **[6, 9, 10, 34]**. In Mali, Department of Traditional Medicine (DMT), current National Institute for Research into Traditional Medicine and Pharmacopoeia (INRMPT), developed two Improved Traditional Medicines for the treatment of eczema. These are the "PSOROPSERMINE" and "MITRADERMINE" pomades, made respectively of *Psorospermum guineense* and *Mitracarpus scaber* **[3]**. Data from this work on *S. ocymoides* species could help expand the range of these natural anti-dermatological products, consequently.

CONCLUSION

This study highlighted medicinal properties and antioxidant, antibacterial and antifungal potential of *S. ocymoides*. The results revealed that local populations living in the rural commune of Dinandougou used this species to treat a large number of diseases and disorders, including dermatological problems (ringworm), malaria and wounds. Phytochemical investigations have shown that extracts of this species are rich in bioactive compounds and are endowed with antioxidant, antibacterial and antifungal properties. As a result, the species could be used for the prevention or management of certain diseases linked to oxidative stress, as well as those linked to bacterial and fungal infections. Lastly, investigations of toxicological properties are needed to enhance the value of this species.

CONFLICT OF INTEREST

Authors declared the absence of any conflict of interest.

AUTHORS' CONTRIBUTIONS

MAK and ST developed the protocol, carried out the survey, the samples collection, the biological tests, analyzed the data, and drafted the manuscript. This work was carried out under the coordination and supervision of MH and ND. All authors read and approved the final version of the manuscript.

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