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Original Research Article

Phytochemical study, antioxidant and antibacterial activities of an Anti-haemorrhoidal medicinal plant recipe used in three towns in Burkina Faso

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Present study aim to carry out an ethnobotanical survey into medicinal plant recipes used in the treatment of hemorroidal pathologies and select the most cite for phytochemical and biological investigation, using ethanolic and hydroethanolic extracts. Phytochemistry study concerned tube test and polyphenolic quantification. Biological one concerned the antioxidant and antibacterial activity. The Folin-Ciocalteu reagent, $AlCl_3$ and vanillic acid were used respectively for the detremination of total phenolics, flavonoids and tanins content. Anti-DPPH*and anti-FRAP were used to evaluate the antioxidant activity and liquid dilution method for the antibacterial activity. 55 medicinal plant recipes were identified with *T. emetica* and *C. sieberiana* the most cited species. Saponosides, flavonoids, tannins and alkaloids were identified in the both extracts. Among the total extracts, *C. sieberiana* gave the high content of phenolic compounds (75.76 ± 0.44 mgEAG/100mg) and flavonoids (0.63 ± 0.36 mgEQ/100mg) respectively with the ethanolic and hydroethanolic extracts on the other hand the best tannin content was observed by the recipe hydroethanolic extract (47.48 ± 3.81 mgEC/100mg). Regarding the antioxidant activity the best anti-DPPH* inhibition percentage was obtained with the hydroethanolic extract of *C.sieberiana* (88.99 %) and the best iron reducing power with the recipe of ethanolic extract (6.52 mmolAA/g). The best Minimum Inhibitory Concentration of 0.78mg/mL was observed on the species of *E. coli* and *K. pneumoniae* with the ethanolic extract of *C. sieberiana*. Also the best Minimum Bactericidal Concentration (25 mg/mL), was observed on the majority of bacterial strains and with all of our extracts. Present findings could partially justify the traditional uses of these plants in the treatment of hemorroidal pathologies.

Keywords: Phytochemistry, Antibacterial, medicinal plant recipes, anti-hemorrhoidal

INTRODUCTION

Haemorrhoidal disease is a worldwide, multifactorial condition and is one of the major problems in anorectal diseases (Kamsu, 2011). Worldwide, its prevalence varies from 4.4 to 86% depending on the population studied (Haas et al., 1984). In the United States, for example, this condition affects 50% of the population and 82.9% of people hospitalised in France (Johanson and Sonnenberg, 1990 ; Dixon et al., 2004 ; Sénejoux, 2010). In Africa, more

specifically in Senegal and Burkina Faso, it accounts for 93.1% of anal lesions and 45.6% of anorectal consultations respectively (Dia et al., 2010 ; Guingané et al., 2014). As far as the management of the disease is concerned, many treatments are offered by modern medicine. Although effective, these treatments are not without risk to human health. They are also inaccessible to the majority of the population (Higuero, 2014). In addition to modern medicine, there is also traditional medicine, which uses recipes based on medicinal plants. According to some authors, over 80% of the world's population and 90% of the inhabitants of Burkina Faso use medicinal plants for their primary healthcare (Zerbo et al., 2007 ; Laleye et al., 2014). These plants contain compounds with several biological activities (Abudunia, 2018). The search for effective biomolecules with fewer iatrogenic effects would be a good alternative. Previous works in markets and villages in central and coastal Cameroon have identified 60 medicinal plants used to treat hemorrhoidal diseases (Dibongo et al., 2015). Equally in Côte d'Ivoire 17 medicinal plants were inventoried (Sidio et al., 2020) with *Alchornea cordifolia* the most cited. Similarly, in Burkina Faso, investigations in the Hauts Bassins (Bobo-Dioulasso) identified 23 medicinal anti- hemorrhoidal diseases plants (Kamboulé et al., 2020) with *Trichilia emetica* vahl most cited. It is true that scientific studies have done on anti-hemorroidal medicinal plants but according to present knowledge, there is little scientific information about the medicinal plant recipes. The general objective of this study is to contribute to the knowledge of medicinal plant recipes used in hemorroidal pathologies treatment by evaluating phytochemical, antioxidant and antibacterial activities. Specifically, it is about :

- inventorying the medicinal plant recipes used in hemorroidal pathologies treatment by the traditional health practitioners of Bobo-Dioulasso, Orodara and Dédougou,
- determine chemical groups in the recipes extracts,
- measuring the total phenolics, flavonoids and tannin content of recipe extracts and their antioxidant power (Anti-DPPH*and anti-FRAP),
- evaluate the ratio of minimum bactericidal concentration (MBC) and minimum inhibitory concentration (MIC) finally to determine if the extracts are bactericidal or bacteriostatic.

MATERIALS AND METHODS

Plant materials

The plant material consisted of *Trichilia emetica* Vahl and *Cassia sieberiana* DC roots collected in the Dindéresso (*Cassia sieberiana* DC) and Péni (*Trichilia emetica* Vahl) classified forest respectively the 14/04/2022 and 03/05/2022 in the company of Botanist Dr Hermann Yempabou OUOBA.The samples were cleaned, dried, pulverised and packed in zip-lock bags.

Bacterial strains

The bacterial material was obtained in the bacteriology and virology laboratory of the Protestant Hospital center in Ouagadougou (SHIPHRA) on hospitalized and non-hospitalized patients.It is composed solely of bacterial isolat : *Escherichia coli (ESBL), Shigella sonnei, Klebsiella pneumoniae Citrobacter freundii, Salmonella typhi, Citrobacter sp, Pseudomonas aeruginosa, Salmonella sp, Klebsiella pneumoniae (ESBL), Escherichia coli* and fermentative name: *Actinobacteria Baumanni.*

Reagents and Solvents

The reagents and solvents used during the manipulations were the following: ascorbic acid, and ferric chloride ($FeCl_3$) (Sigma), aluminum chloride ($AlCl_3$) (Sigma-Aldrich, Germany), quercetin (Sigma), 2,2-diphenyl-1-picrylhydrazyl (DPPH) (Sigma,USA), Folin-Ciocalteu reagent (sigma,USA), gallic acid(Sigma), sodium carbonate(Sigma, Germany), ethanol, sodium chloride (Sigma,Germany), glycerol, distilled water and bleach (Labioca,Burkina),concentrated hydrochloric acid (HCl) (Sigma,USA), magnesium (Mg) (Sigma), vanilic acid (Sigma), trichloroacetic acid, potassium hexacyanoferrate [K3Fe (CN6)] (Sigma), catechol(Sigma).

Ethnobotanical survey

The ethnobotanical survey was carried out in three towns in Burkina Faso : Bobo-Dioulasso, Dédougou and Orodara with traditional healers and dozos in the month of October 2021 to January 2022. Pre-established survey forms were used to interview traditional health practitioners. This was semi-structured interview with a single passage. The data collected concerned the respondent and the recipes used to treat haemorrhoidal disease.

Extraction

A mass 15 g of powder from each sample was mixed with 150 mL of ethanol or ethanol-water (20 ; 80 v/v) and stirred for 48h. The macerate was filtered and the dry extracts obtained were used for phytochemical and biological studies.

Phytochemical analysis

Phytochemical screening

The purpose of general characterisation tests is to reveal the major groups of secondary metabolites present in plant extracts (Ciulei, 1982). These tests were carried out on macerated extracts:
-The reaction with $FeCl_3$ is used for tannins and
-The Shibata test for flavonoids
-Dragendorff test (potassium tetra iodo bismuthate) for alkaloids ;

-Foam test for saponosides.

Quantification of polyphenols

Total phenolic acids assay

The total phenolic content was estimated using the Folin Ciocalteu reagent (Singleton et al., 1999). Briefly, 400 µL of RCF (0.2%) plus 420 µL of NaCO3 were added to 100 µL of extract at a concentration of 1 mg/ml. The mixture was incubated for 2 h against a blank consisting of 100 µL of extract, 400 µL of 80% ethanol and 420 µL of NaCO3. For each sample, reading was performed in triplicate using a spectrophotometer at 760 nm against a calibration curve (y = 2.4343x + 1.118 with R^2 = 0.99). The results were expressed in mg Gallic Acid Equivalent (mg GAE) per 100 mg of dry extract.

Total flavonoids assay

Total flavonoid content was estimated using aluminium trichloride (Lamien-Meda et al., 2008). A volume of 500 µL of $AlCl_3$ (2%) prepared in ethanol was mixed with 500 µL of extract (0.1mg/mL) and incubated for 15 min. Spectrophotometer readings were taken at 415 nm against a blank containing 500 µL extract and 500 µL ethanol. Readings were taken in triplicate per sample against a standard curve (y= 34.931x+ 0.0189 with R^2 =0.9981) obtained from successive dilutions of a quercetin stock solution (500 mg/L to 1 mg/L). Results were expressed as mg Quercetin Equivalent (mgEQ) per 100 mg dry extract.

Total tannin determination

Total tannin content was estimated using the method describ by Bangou et al. (2011), briefly, 1000µl of vanillin solution (c=0.06mg/ml) was added to 200µl of each extract and the mixture was incubated for 20min. A blank consisting of 1000µl vanillin anbd 200µl 80% ethanol was also added. Absorbance was measured at a wavelength of 500nm. Three tests were carried out for each sample. A stock solution of catechol was used as the reference standard to establish the calibration curve (y = 0.1239x + 0.0005; R^2 =0.9986) and the contents were expressed as mgEC/100mg dry matter.

Assessment of antioxidant activity

Free radical Scavening activity

Reductive activity by the DPPH method was carried out according to the protocol of Velázquez et al. (2003). Briefly, a mother extract (1 mg/mL) was diluted 1:10 in 80% ethanol and 400 µL (0.1 mg/mL) was added to 800 µL DPPH (0.2 mg/mL) in three test tubes incubated for 15 min in the dark against a blank consisting of 800 µL DPPH (0.2 mg/mL) and 400 µL 80% ethanol. Absorbances were read using a spectrophotometer at 517 nm.

Iron-reducing power

The FRAP (Ferric Reduccing Antioxidant Power) method is based on the ability of extracts to reduce ferric ion (Fe^{3+}) to ferrous ion (Fe^{2+}) (Hinneburg et al., 2006). The method was carried out as follows : 0.5 mL of an aqueous solution of each extract (0.1 mg/mL) was mixed with 1.25 mL of phosphate buffer (pH=6.6 ; 0.2) and 1.25 mL of 1% potassium hexacyanoferrate [K_3 Fe (CN_6)]. The resulting solution was placed in a water bath, after which 1.25 mL of 10% trichloroacetic acid was added, incubated for 30 min and then centrifuged at 200 rpm for 10 min ; R^2 =0.9976) obtained from ascorbic acid.To 500 µL of distilled water was added 100 µL of 0.1% FeCl iron trichloride$_3$ and 500 µL of the freshly prepared solution in three test tubes incubated for 20 min against a blank consisting of 1000 µL of distilled water and 100 µL of $FeCl_3$.Absorbances were read using a spectrophotometer at 700 nm against a standard ($y=6.1.10^{-2}$ x + 0.0037) obtained from ascorbic acid.

Study of the recipe's antibacterial properties

Preparation of the inoculum

bacterial strains stored in cryotubes were inoculated onto MH agar plates incubated for 24 hours at 37°C in order to obtain young isolated strains which were collected with a pasteur pipette, homogeneously in 10 mL of MH broth.

Determination of the MIC (minimum inhibitory concentration)

A concentration 50 mg/mL of initial volume of each recipe extract was prepared. From the stock solution, a series of dilutions was carried out using a geometric progression of reason 2 to obtain concentration ranges from 50mg/ml to 0.048mg/ml according to Arias et al. (2004).

The antibacterial tests were carried out using the liquid dilution method (Kouadio et al., 20 15). 100 µL of sterile MH broth was placed in the 96 wells of the plate, and 100 µL of the extract from the recipe was added to the first well of each line, which was then diluted up to the eleventh well, the no-growth control well. In addition, 100 µL of the bacterial strain inoculum is added to all 12 wells in the corresponding line except for the eleventh well. The twelfth is the growth control after the plate has been incubated for 24 hours at 37°C. After 24 hours, the reading enabled Sto determine the MIC (minimum inhibitory concentration) of the extract where there was no growth observed by the naked eye (Olivia et al., 2003).

Determination of the MBC (minimum bactericidal concentration)

This was carried out using the method developed by *Zongo* et al. (2011).

Statistical analysis

Statistical analysis : All assays were carried out in triplicates and results are expressed as Means ± Standard Deviation (SD) calculated with Excel 2016.

RESULTS AND DISCUSSION

ethnobotanical survey data

The ethnobotanical survey enabled us to identify 55 medicinal plant recipes in the three study towns. The distribution of the species used in the recipes showed the following proportions : 48, 34.66, 10.67 and 6.67% respectively for recipes composed of 1, 2, 3, 4 or more species. These results shows that one or more species, alone or in combination, can be used to treat haemorrhoids disease. According to the study of Said et al. (2022), the therapeutic effect of recipes composed of several plant species could be due to a synergy of action between the different plants.

The ethnobotanical survey identified 45 medicinal plant species belonging to 28 botanical families, of which *Trichilia emetica* Vahl and *Cassia sieberiana DC* were the most frequently cited. Of the various families, Meliaceae (29.55%), followed by Fabaceae (24.81%) were the most represented. The same observation was made by Kamboulé et al. (2020) who found 36.11% and 5% respectively.

Parts of plants used in the recipes

Figure 1 illustrates the distribution of the different organs of medicinal plant species used by traditional healers. It can be seen that all parts of the plant are involved in the preparation of recipes, but this varies from city to city. Analysis of the results shows that roots followed by barks are used in the majority of cases, respectively 38.1%, 50.9% and 27.78% in Bobo-Dioulasso, Orodara and Dédougou for the roots and 46.82% and 22.22% in Dédougou and Bobo-Dioulasso for the barks. According to Zerbo et al. (2011), these parts are where secondary metabolites are stored. Indeed, the high use of bark could be due to the richness of this organ in chemical substances since the bark is the communication route between the roots and the leaves (Dongock et al., 2018).

Distribution according to the methode of preparation the Recipes

Figure 2 shows the distribution of recipe preparation methods. We note that five methods - decoction, infusion, maceration, powder and incineration - are used to prepare the recipes. The analysis shows that decoction was the method most used in the three city, with 73.91%, 65.46% and 68.75% respectively in Bobo-Dioulasso, Orodara and Dédougou. Similar investigations by Sawadogo et al. (2022) and Lema et al. (2022) also mentioned decoction as the most widely used preparation method, with 54.29% and

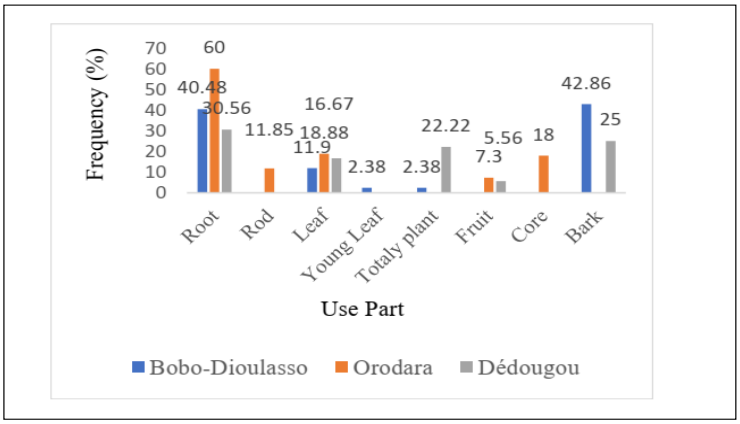

Figure 1: Frequency of plant parts useds

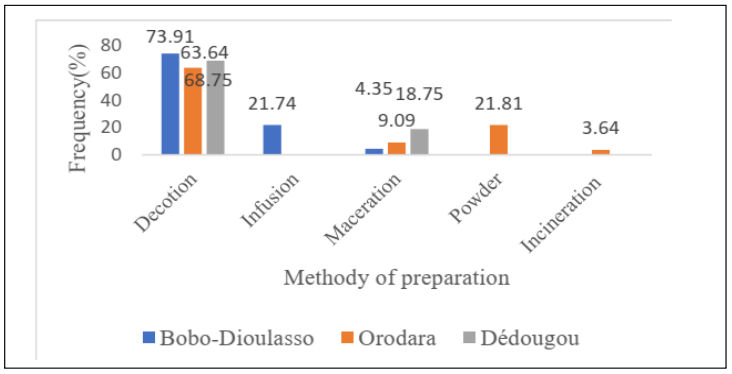

Figure 2 : Distribution according to the method of preparation

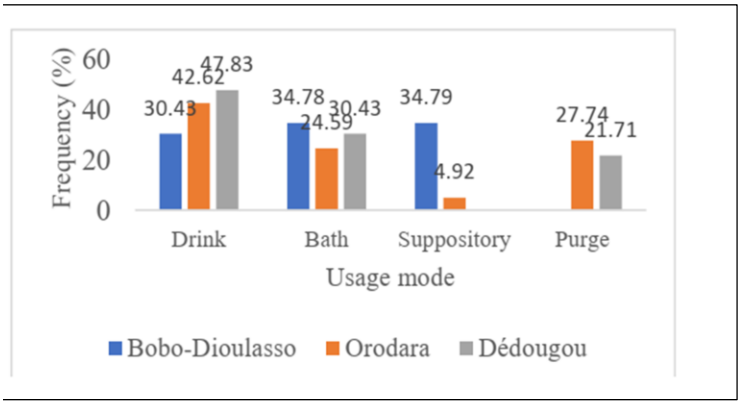

Figure 3 : Method of administration

70% respectively. According to Salhi et al. (2010), decoction allows more active principles to be collected and attenuates or cancels out the toxic effect of certain recipes.

Distribution of revenue administration the methods

Figure 3 shows the distribution of revenue administration

Table 1 : Phytochemical screening results

Types of extracts		Chemical groups			
		Saponosides	Flavonoids	Tannins	Alkaloids
Ethanol extracts	*C. siberiana*	+	+	+	+
	T. emetica	+	+	+	+
	Recipe	+	+	+	+
Hydroethanol extracts	*C. siberiana*	+	+	+	+
	T. emetica	+	+	+	+
	Recipe	+	+	+	+

(Recipe) : mixture of powder of the two species ; (+) : presence

Table 2. Quantification of total polyphenols

Types of extracts		Content of polyphenolic compounds (mg Eq/100mg extract)		
		Total phenolics (GA)	Total flavonoids (Q)	Total tannins (C)
Ethanol extracts	*C. sieberiana*	75.76 ± 0.44 ^{a}	0. 42 ± 0.0 ^{a}	7.66 ± 0.88 ^{d}
	T. emetica	56.48 ± 0.44 ^{d}	0.34 ± 0.02 ^{a}	7.13 ± 1.23 ^{d}
	Recipe	56.25 ± 0.53 ^{d}	0.23 ± 0.43 ^{a}	28.65 ± 3.51 ^{b}
Hydroethanol extracts	*C. sieberiana*	74.76 ± 0.44 ^{b}	0.63 ± 0.36 ^{b}	11.97 ± 3.98 ^{c}
	T. emetica	54.92 ± 0.64 ^{e}	0.54 ± 0.18 ^{b}	8.20 ± 1.23 ^{e}
	Recipe	62.53 ± 0.68 ^{c}	0.24 ± 0.02 ^{a}	47.48 ± 3.81 ^{a}

Values are mean ± SD (n = 3). Different letters in the same column indicate significant difference ($p < 0.05$)

methods. We note four methods of revenue administration in the three towns. We note that drinking (42.62% ; 47.83%) was the mode most used in Orodara and Dédougou. This method was also mentioned by other authors such as Jazy et al. (2017) ; Sawadogo et al. (2022) ; Lema et al. (2022) (77.14% and 47%) in their study as being the most recommended by traditional practitioners.

Phytochemical screening

These tests were carried out to highlight the presence of certains groups of chemical compounds in the extracts (Table 1). This study revealed that all our extracts contained Saponosides, flavonoids, tannins and alkaloids. Other previous studies have also revealed the presence of these chemical groups in these species (Djoupo et al., 2015; Mshelia et al., 2017).

Total phenolic, flavonoid and tannin

The total phenolic content of the different extracts is shown in Table 2. They ranged from 54.92±0.64 mgEAG/100mg to 75.76±0.44 mgEAG/100mg. The highest content was obtained with the ethanolic extract of *C. Sieberiana* DC (75.76±0.44 mgEAG/100mg) and the lowest with the hydro-ethanolic extract of *T. emetica* Vahl (54.92±0.64 mgEAG/100mg).

Total flavonoid levels ranged from 0.23±0.43 mgEQ/100mg to 0.63±0.36 mgEQ/100mg. The highest content was obtained with the hydroethanolic extract of *C. Sieberiana* DC (0.63±0.36 mgEQ/100mg) and the lowest with the ethanolic extract of the recipe (0.23±0.43 mgEQ/100mg).

Total tannin levels varied from 7.13±1.23 mgEC/100mg to 47.48±3.81 mgEC/100mg. The highest content was obtained with the hydro-ethanolic extract of the recipe (47.48±3.81 mgEC/100mg) and the lowest with the ethanolic extract of *T. emetica* Vahl (7.13±1.23 mgEC/100mg). The results show that polyphenolic compounds are abundant in the extracts. According to Ghedadba et al. (2014), the high polyphenol content is related to the high solubility of phenols in polar solvents.

Evaluation of antioxidant activities

The antioxidant power of the extracts was assessed namely the method for trapping the DPPH radical and the reducing power of iron by the FRAP method (Table 3). For the DPPH radical trapping method, the inhibition percentages varied from 74.99 to 88.99.the highest inhibition percentages was obtained with the hydroethanol extract of *C. Siberiana* (88.99%) and the lowest with the ethanol extract of the same species (74. 99%).

For the FRAP reducing power method, the activity of extracts varied from 3.81± 0.13 mmol EAA/g to 6.52± 0,19 mmol EAA/g. The best activity was obtained with the ethanolic extract of the recipe and the lowest with the hydroethanolic extract of *T.emetica* with respectively 6.52±

Table 3. Antioxidant activity results

Type of extract		DPPH (I%) 0.1mg/ml	FRAP (mmol EAA/g of extract)
Extracts éthanolic	*C. siberiana*	74.99	4.98±0.06 [c]
	T. emetica	76.99	4.43±0.06 [b]
	Recette	83.99	6.52±0.19 [e]
Extracts hydroéthanolic	*C. siberiana*	88.99	4.78±0.17 [c]
	T. emetica	79.98	3.81±0.13 [a]
	Recette	84.98	6.20±0.3 [d]

Values are mean ± SD (n = 3). Different letters in the same column indicate significant difference ($p < 0.05$)

Table 4 : Minimum Inhibitory Concentration values for recipes (mg/mL)

	Ethanol extracts			Hydroethanol extracts		
Bacterial strains	***C. sieberiana***	***T. emetica***	**Recipe**	***C. sieberiana***	***T. emetica***	**Recipe**
E. coli (ESBL) (2457)	0.78	12.5	3.125	1.56	12.5	1.56
E. coli (ESBL) (2704)	3.125	12.5	12.5	12.5	12.5	1.56
S. sonnei (1264)	0.78	6.25	6.25	1.56	12.5	3.125
E. coli (ESBL) (2455)	0.78	6.25	6.25	1.56	12.5	3.125
K. pneumoniae (2527)	0.78	12.5	12.5	3.125	6.25	6.25
C. freundii (2525)	12.5	12.5	12.5	3.125	6.25	6.25
Salmonella sp (1148)	12.5	12.5	12.5	3.125	6.25	6.25
A. Baumanni (2532)	12.5	12.5	12.5	3.125	6.25	6.25
K. pneumoniae (ESBL) (2582)	12.5	12.5	12.5	3.125	6.25	6.25
P. aeruginos (982)	1.56	25	3.125	3.125	12.5	6.25
E. coli (565)	1.56	25	3.125	3.125	12.5	3.125
K. pneumoniae (273)	1.56	25	50	3.125	12.5	50
E. coli (714)	12.5	12.5	12.5	3.125	6.25	6.25
S. typhi (176)	1.56	12.5	3.125	3.125	12.5	3.125
K. pneumonia (273)	12.5	12.5	12.5	3.125	6.25	6.25
P. aeruginos (93)	12.5	12.5	12.5	3.125	6.25	6.25
E. coli (241)	6.25	3.125	3.125	6.25	3.125	3.125
Citrobacter sp (1082)	6.25	1.56	6.25	6.25	1.56	6.25

Recipe) : mixture of powder of the two species

0.19 mmol EAA/g and 3.81± 0.13 mmol EAA/g. The extracts showed a good percentage of radical inhibition ranging to 0.1 mg/ml lower than that of ascorbic acid with a concentration of 0.01 mg/ml for 96.44%. This shows that these extracts could be powerful inhibitors of the free radicals that cause inflammation and pain, hence their use in the treatment of haemorrhoidal disease.

Antibacterial activity

Minimum Inhibitory Concentration

The minimum inhibitory concentration (MIC) was determined for Gram-negative bacteria producing β-lactamase (ESBL) or not ESBL. These results are shown in Table 4. The MIC of all extracts on the different bacterial strains ranged from 0.78mg/mL to 12.5mg/mL. The best Minimum Inhibitory Concentration of 0,78mg/mL was observed on the species of *E. coli* and *K. pneumoniae* with the ethanolic extract of *C. sieberiana*. These results indicate bacterial activity of all extracts on bacteria.

Minimum Bactericidal Concentration

The MBC is the Minimum Bactericidal Concentration. 50 mg/mL on the different β-lactamase (BLSE) producing or non-β-lactamase producing bacteria in Table V. This concentration of extracts varied from 25 mg/mL to 50 mg/mL on the different β-lactamase (BLSE) producing or non-β-lactamase producing bacteria in Table (5).

Bacteriostatic and Bactericidal

The MBC/MIC ratio is used to determine whether the extract is bactericidal or bacteriostatic and if this ratio is equal to 1 or 2, we have a bactericidal effect and when the ratio is greater than or equal to 4 we have a bacteriostatic effect (Bangou, 2012). The MBC/MIC ratio of all our extracts varies from 1 to 64. In view of our results, we can say that the extracts of both species had a bactericidal and bacteriostatic effect on the bacterial strains(Table 6). Specifically, the hydroethanol extracts of *C. sieberiana* and the recipe were bactericidal on *E. coli (ESBL) (2704).* The

Table 5. Minimum Bactericidal Concentration values for recipes (mg/mL)

	Ethanol extracts			Hydroethanol extracts		
Bacterial strains	***C. sieberiana***	***T. emetica***	**Recipe**	***C. sieberiana***	***T. emetica***	**Recipe**
E. coli (ESBL) (2457)	25	50	25	25	50	25
E. coli (ESBL) (2704)	25	50	25	25	50	25
S. Sonnei (1264)	25	25	25	25	25	25
E. coli (ESBL) (2455)	25	25	50	25	25	50
K. pneumoniae (2527)	50	50	50	50	50	50
C. freundii (2525)	50	50	50	50	50	50
Salmonella sp (1148)	50	50	50	50	50	50
A. Baumannie (2532)	50	50	50	50	50	50
K. pneumoniae (ESBL) (2582)	50	50	50	50	50	50
P. aeruginos (982)	25	50	25	25	50	25
E. coli (565)	25	50	50	25	50	25
K. pneumoniae (273)	25	50	50	25	50	50
S. typhi (176)	25	50	25	25	50	50
E. coli (714)	50	50	50	50	50	50
K. Pneumoniae (273)	50	50	50	50	50	50
P. aerugino (93)	50	50	50	50	50	50
E. coli (241)	50	50	50	50	50	50
Citrobacter sp (1082)	50	50	50	50	50	50

Recipe) : mixture of powder of the two species

Tables 6. Value of the MBC/MIC ratio

	Ethanol extracts			Hydroethanol extracts		
Species	***C. sieberiana***	***T. emetica***	**Recipe**	***C. sieberiana***	***T. emetica***	**Recipe**
E. coli (ESBL) (2457)	32	4	8	16	4	16
E. coli (ESBL) (2704)	8	4	2	2	4	16
S. Sonnei (1264)	32	4	4	16	2	8
E. coli (ESBL) (2455)	32	4	8	16	2	16
K. pneumoniae (2527)	64	4	4	16	8	8
C.freundii (2525)	4	4	4	16	8	8
Salmonella sp (1148)	4	4	4	16	8	8
A. Baumannie (2532)	4	4	4	16	8	8
K. pneumoniae (ESBL) (2582)	4	4	4	16	8	8
P. aerugino (982)	16	2	8	8	4	4
E. coli (565)	16	2	16	8	4	8
K. pneumoniae (273)	16	2	1	8	4	1
S. typhi (714)	16	4	8	8	4	16
E. coli (714)	4	4	4	16	8	8
K. Pneumoniae (2582)	4	4	4	16	8	8
P. aerugino (93)	4	4	4	16	8	8
E. coli (241)	8	16	16	8	16	16
Citrobacter sp (1082)	8	32	8	8	32	8

(Recipe) : mixture of powder of the two species
MBC/MIC ≤ 4 mg/mL is ***Bactericidal****. ; MBC/MIC ≥ 4 mg/mL is* ***Bacteriostatic****.*

ethanolic extract of *T. emetica was* also bactericidal against *P. aerugino (982), Escherichia coli (565)* and *Klebsiella pneumoniae* (273). The ethanolic and hydroethanolic extracts in the recipe are bactericidal against *K. pneumoniae* (273). The bactericidal effect of these extracts could be explained by their richness in polyphenols, more specifically tannins, because according to quantification tests, these extracts contain higher levels of total tannins. According to Bakasso, (2009), phenolic compounds, particularly tannins, are susceptible to polymerisation in space during oxidation reactions, and this could also be a toxicity factor for microorganisms.

CONCLUSION

Ethnobotanical surveys of medicinal plant recipes used in the treatment of hemorroidal diseases in the urban areas of Bobo-Dioulasso, Orodara and Dédougou allowed us to identify 55 medicinal plant recipes with *T. emetica* and *C.*

sieberiana the recipe selected. Phytochemical investigations revealed saponosides, flavonoids, tannins and alkaloids in the extracts. From the quantification tests, the best total phenolic contents were obtained with the ethanolic and hydroethanolic extracts of *Cassia sieberiana* DC 75.76 ± 0.44 mgEAG/100mg extract and 74.76 ± 0.4 mgEAG/100mg respectively. With regard to total tannins, the best content was obtained with the hydroethanolic extracts of the recipe, be 47.48 ± 3.81 mgEC/100mg. With regard to antioxidant activity, the best reducing power iron was obtained with the ethanolic extract of the recipe ie 6,52 mmol EAA/g and the best inhibition percentage was obtained with the hydroethanolic extracts of *C. sieberiana*, i.e 88.99% using the DPPH radical inhibition method. In terms of antibacterial activity, the best MIC was obtained by the hydroethanolic extract of *C. sieberiana* DC, i.e 0.78 mg/mL, and the best BMC was obtained by all the extracts on different strains, be 25 mg/mL. The best bactericidal activity was observed with the ethanolic extract of *T. emetica* Vahl and the best bacteriostatic activity with the hydroethanolic extract of both species. These different phytochemical and biological results could justify the use of these plant recipes in the treatment of hemorrhoidal diseases. Consequently, the recipe for *C. Sieberiana* DC and *T. emetica* Vahl could be the subject of in-depth analysis with the aim of scientifically validating its traditional use against hemorrhoidal diseases.

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