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Phytochemical screening and evaluation of antiradical and antimicrobial activities of *Annona senegalensis* and *Detarium microcarpum* leaves used in Benin to treat urinary infections

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Abstract

The challenge of resistance to synthetic antimicrobials necessitates new alternative solutions. Medicinal plants are rich in active compounds with various medicinal properties that can be used in treatment of urinary infections. This work aims to highlight the value of *Annona senegalensis* and *Detarium microcarpum* by evaluating phytochemical and antimicrobial potentials of their leaf extracts. Qualitative and quantitative analyses were carried out using color and precipitation reactions and spectrophotometric assays. The antiradical activity was evaluated using the DPPH test, and the antimicrobial activity was assessed by the agar diffusion method. Phytochemical screening showed that both plants contain anthraquinones, catechic tannins, coumarins, flavonoids, reducing compounds, sterols and terpenes. Hydroethanolic extract of *Detarium microcarpum* and *A. senegalensis* showed IC_{50} inhibition percentages of 0.20 mg/mL and 0.22 mg/mL respectively. Regarding antimicrobial activity, the hydroethanolic leaf extracts of *D. microcarpum* and A. senegalensis were found to be bactericidal and fungicidal against the tested bacteria and molds. This extract showed more pronounced antimicrobial activity than amoxicillin and ciprofloxacin, which are synthetic antimicrobials. Diversity of secondary metabolites and notable antimicrobial activity of *D. microcarpum* and *A. senegalensis* may justify their use in traditional medicine for treating urinary infections in Benin.

Keywords: Urinary infections; Medicinal plants; Secondary metabolites; Antiradical; Bactericidal; Fungicidal

1. Introduction

Benin, like many West African countries, uses medicinal plants to treat diseases in traditional medicine [1]. This medicine is based on empirical knowledge passed down from generation to generation. Plants are valuable resources for the vast majority of rural populations, with more than 80% using them for their healthcare needs [2], [3]. Urinary tract infections are common ailments affecting various populations worldwide. These infections, primarily caused by bacteria, affect the urinary tract and can manifest in different forms such as cystitis, urethritis and pyelonephritis [4]. They represent a significant public health issue, being second most common infectious disease after respiratory tract infections [5], [6]. They can have severe consequences, particularly for pregnant women or patients with urinary tract abnormalities or predisposing factors such as diabetes or immunodepression [7]. Women, in particular, are more susceptible to these infections due to anatomical proximity of the urethra and anus, facilitating bacterial entry into the urinary tract [8], [9]. Living conditions, limited access to quality medical care, and certain behaviors contribute to high incidence of these infections. They are mainly caused by bacteria such as *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Staphylococcus saprophyticus*, and *Proteus mirabilis* [4],[10],[11],[12]. *Annona senegalensis* and *Detarium microcarpum*, from the Annonaceae and Fabaceae families respectively, are among plants heavily used in

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traditional medicine in Benin to treat urinary tract infections. They are used in traditional medicine to treat various ailments, including infections, fever, pain, chickenpox, tuberculosis, diabetes, malaria, stomachaches, dysentery, sexually transmitted diseases, and diarrhea [13], [14],[15]. With the increase in bacterial resistance and the relatively high cost of conventional medicines, scientific exploration of medicinal plants becomes an ideal alternative. This work aims to highlight *Annona senegalensis* and *Detarium microcarpum* through phytochemical characterization and evaluation of their antiradical and antimicrobial activities of their leaves.

2. Materials and Methods

2.1. Materials

2.1.1. Plant material

The plant material consists of leaves from *Annona senegalensis and Detarium microcarpum* collected in northern Benin. The plants were identified and voucher specimens prepared by the botanist Professor Hounnankpon Yedomonhan of the Benin National Herbarium where they were deposited under the specimen numbers YH754/HNB and YH756/ HNB for *Annona senegalensis and Detarium microcarpum* respectively.

2.1.2. Chemicals

Methanol, Folin–Ciocalteu reagent, 2,2-diphenyl-1-picrylhydrazyl (DPPH), quercetin, gallic acid, aluminum chloride, potassium acetate and sodium acetate were purchased from Sigma-Aldrich. All reagents and chemicals were analytical grade.

2.1.3. Microbial strains

The microbial material consists of sixteen (16) bacterial strains such as *Staphylococcus aureus*, *Escherichia coli*, *Escherichia coli* ATCC 25922, *Streptococcus* D, *Klebsiella pneumoniae*, *Streptococcus* D and *Candida albicans* MHMR. The bacterial strains are clinical isolates obtained from urine and semen.

2.2. Methods

2.2.1. Annona senegalensis and *Detarium microcarpum* leaves pretreatment

After identification of plant samples at National Herbarium of Benin, they were dried in laboratory until their plant mass stabilized before being ground into powder.

2.2.2. Plant extracts

The extraction was made with ethanol and hydroethanolic under ultrasounds. Briefly, 10 g of powdered biomass were mixed with 100 mL solvent and sonicated for two hours at 50 °C with Bandelin (Sonorex Digitech device). Further, all extracts were filtered through Whatman No.1 filter paper and concentrated under vacuum (Buchi R215, heating bath B-491, rotation 280 rpm, vacuum controller V-850 of 290 mbar) at (50±1) °C. The residues were dried to constant weights and stored in the darkness at 4 °C to avoid degradations until use [16], [17].

2.2.3. Preliminary phytochemical screening

The secondary metabolites of *Annona senegalensis* and *Detarium microcarpum* were identified through specific color reactions and precipitation reactions unique to each secondary metabolite, as summarized in Table 1 [18],[19],[17].

Table 1 Methods for the identification of secondary metabolites of *Annona senegalensis* and *Detarium microcarpum*

Secondary metabolites	Chemical test
Alkaloids	Mayer's test and Dragendroff's test
Anthocyanes	test with hydrochloric acid and ammonia
Anthraquinones	Borntranger's test
Coumarins	365 nm , fluorescence test
Flavanoids	Shibita's reaction test

Tannins	stiasny test, ferric chloride and sodium acetate test
Saponins	Frothing test
Leuco anthocyanins	Bate-Smith and metcalf
Mucilages	flaky test
Cyanogenic derivatives	picric acid test
Reducing compound	Fehling's test
Sterols and terpenes	Liebermann-Burchard's test)

2.2.4. Determination of phenolic compounds

- **Total phenol content**: Total phenolic content was determined using the Folin-Ciocalteu colorimetric method Lupoae *et al*.[20] with some modifications. This method consisted of using a mixture of phosphotungstic and phosphomolybdic acids, which were reduced during the oxidation of phenols into a mixture of tungsten blue oxide and molybdenum. Finally, absorbance was measured at 760 nm using a spectrophotometer (with Infinite 200 PRO-Tecan microplate) and total phenol content are expressed in micrograms of gallic acid equivalence per milligram of extract (µgGAE/mg Ex) [20],[21],[22].
- **Total flavonoids content**: Method of aluminum trichloride ($AlCl_3$) was used to quantify total flavonoids. This technical was based on formation of aluminum complex flavonoids. Absorbance was read at 415 nm using a spectrophotometer (Infinite 200 PRO-Tecan microplate) and Total flavonoid content are expressed in micrograms quercetin equivalence per milligram of extract (µgQE/mg Ex)[23].
- **Condensed tannin content**: Condensed tannins are measured using Butanol-HCl method. Reaction medium is composed of 0.5 mL of extract, 3 mL of butanol-HCl (95/5), and 0.1 mL of a ferric solution (2% ferric ammonium sulfate diluted in 2N HCl). Samples are incubated in a boiling water bath for 60 minutes. Absorbance is measured at 550 nm, and results are expressed in leucocyanidin equivalents, according to following formula: T (mgLE/gEx) = (A x 78.26 x DF); A: is absorbance recorded at 550 nm; DF: dilution factor. The dilution factor is equal to 1 if the extract is prepared at 200 mg in 10 mL of solvent and measured absorbance is less than 0.6 [24].

2.2.5. Antiradical activity

Antiradical activity was evaluated using 2,2-diphenyl-1-picrylhydrazyl (DPPH) method. The principle of this method is based on measurement of free radical scavenging in a DPPH solution. This scavenging is visualized by disappearance of purple color of DPPH. Samples are left in dark for one hour and absorbance is measured at 517 nm [23],[17]. The percentage of radical scavenging was determined using formula : $P=\frac{Aw-As}{Aw}*100$. where As: Sample absorbance; Aw: Absorbance of white.

2.2.6. Antimicrobial activity

- **Sensitivity test:** A bacterial pre-culture (1 colony in 1 mL of liquid Mueller-Hinton) from previous day is diluted to obtain a turbidity of 0.5 on McFarland scale. This bacterial suspension (1000 µL) was introduced into Petri dishes containing Mueller-Hinton agar (Bio Rad, France). Using a perforator, 6 mm diameter paper discs were made. The sterile discs are placed, under aseptic conditions, on bacterial culture plates. On placed discs, 30 µL of the extract to be tested are inoculated under aseptic conditions. For each extract, experiment is duplicated along with a negative control. The plates are then left for 30 minutes at room temperature before being incubated at 37°C in oven for 48 hours before measuring inhibition diameters [25].
- **Determination of minimum inhibitory, bactericidal, and fungicidal concentrations**: Minimum inhibitory, bactericidal, and fungicidal concentrations were determined by microdilution method using iodonitrotetrazolium as an indicator of microbial strain viability [26].

3. Results and discussion

3.1. Secondary metabolites in *Detarium microcarpum* and *Annona senegalensis* leaves

Secondary metabolites identified in the leaves of *Detarium microcarpum* and *Annona senegalensis* are listed in Table 2. The results of preliminary phytochemical screening revealed presence of catechin tannins, flavonoids, anthraquinones, coumarins, reducing compounds, sterols, and triterpenes in *Detarium microcarpum and Annona*

senegalensis leaves. However, mucilages and saponosides were identified only in the leaves of *Detarium microcarpum*, while anthocyanins were found only in the leaves of *Annona senegalensis*. The results of previous studies by Loubaki *et al*.[27] in Niger, Saxena *et al*.[28] and Ebi *et al*.[29] in Nigeria, Akabassi *et al*.[30] in Benin and Hamadou *et al*.[31] in Mali on the leaves of *Detarium microcarpum* corroborate our findings. However, Dembele *et al*.[32] noted absence of tannins in *Detarium microcarpum* from Mali, while tannins were identified in sample from Benin. Similarly, Abdullahi *et al*.[33] reported presence of alkaloids in species from Nigeria, which are absent in the leaves of *Detarium microcarpum* collected in Benin. As for the *Annona senegalensis* leaves collected in Mali, Traoré *et al*.[15] reported presence of alkaloids in their sample, whereas alkaloids were not identified in species from Benin. Yakubu *et al*.[34](2021) noted the absence of anthraquinones in Nigerian species, which were identified in Benin species. However, results of Diallo[35] work with sample from Mali are consistent with our findings. Mbaya *et al*.[36] observed the absence of tannins and anthraquinones in the Nigeria sample, which were identified in our work. The variation in secondary metabolites observed in our samples compared to previous studies could be related to harvest period, soil nature, or climatic factors [37],[38]. The diversity of secondary metabolites in these plants, particularly tannins, flavonoids, coumarins, anthraquinones, sterols, and triterpenes, could explain their use in treating various ailments [39], [40], [41].

Table 2 Phytochemical composition of *Detarium microcarpum* and *Annona senegalensis* leaves

Secondary metabolites	***Detarium microcarpum***	***Annona senegalensis***
	Observation (Present/Absent)	
Alkaloids	Absent	Absent
Anthocyanins	Absent	Present
Anthraquinones	Present	Present
Catechic tannins	Present	Present
Coumarin	Present	Present
Cyanogenic derivatives	Absent	Absent
Flavonoids	Present	Present
Leucoanthocyanins	Absent	Absent
Mucilages	Present	Absent
Reducing compounds	Present	Present
Saponosides	Present	Absent
Sterols and Terpenes	Present	Present

3.2. Phenolic compound content of *Detarium microcarpum* and *Annona senegalensis* leaves

Table 3 presents phenolic compound contents of hydroethanolic extract of *Annona senegalensis* and *Detarium microcarpum* leaves. For *Detarium microcarpum* leaves, phenol content is 94.434 mg GAE/g Ex and 91.99 mg GAE/g Ex for *Annona senegalensis*, with a flavonoid content of 18.419 mg QE/g Ex and 21.25 mg QE/g Ex, respectively, for *Annona senegalensis* and *Detarium microcarpum*. As for tannin content, it is 0.986 mgTAE/gEx for *Detarium microcarpum* and 2.649 mgTAE/gEx for *Annona senegalensis*. For *Detarium microcarpum* species from Benin, Akabassi *et al*.[30] obtained a phenol content of 46.79 mg GAE/g Ex. However, in Niger, Hamadou *et al*.[31] and Lawaly *et al*.[42] found phenol contents of 105 mg GAE/gEx and 40.0 mg GAE/gEx, respectively. Regarding total flavonoid content, hydroethanolic extract of *Annona senegalensis* leaves contains 18.419 mg Q E/g Ex and 21.25 mg Q E/g Ex for *Detarium microcarpum*. Hamadou *et al*.[31] obtained a content of 23.94 mg Q E/g Ex from *Detarium microcarpum* leaves. In contrast, Akabassi *et al*. [30] obtained a content of 30.29 mg Q E/g Ex in Benin, and David *et al*.[43] obtained a content of 30.5 mg Q E/g Ex from a sample in Nigeria. This higher content compared to our results. The variation in phenolic compound content observed in our samples compared to previous studies could be related to harvest period, soil nature, or climatic factors [44],[45].

Table 3 Content of phenolic compounds

Ethanol extract	***Detarium microcarpum***	***Annona senegalensis***
Total phenol content (mg EAG/gEx)	94.43	91.99
Total flavonoid content (mg EQ/g Ex)	21.25	18.419
Condensed tannin content (mg EL/g Ex)	0.986	2.649

3.3. DPPH free radical scavenging assay

3.3.1. Detarium microcarpum

The DPPH· radical scavenging rate as a function of concentrations of hydroethanolic extract of *Detarium microcarpum* leaves is shown by the curve in Figure 1. A progressive increase in the scavenging percentage is observed as concentration of hydroethanolic extract of *Detarium microcarpum* leaves increases, reaching almost 100% before starting to decrease. From this curve, concentration of hydroethanolic extract of *Detarium microcarpum* leaves required to scavenge 50% of DPPH radical is 0.20 mg/mL. These results are consistent with those of Akabassi *et al*.[30], who showed that *D. microcarpum* has very interesting antiradical activity. David *et al*.[43]similarly observed pronounced activity in *Detarium microcarpum* leaves collected in Nigeria.

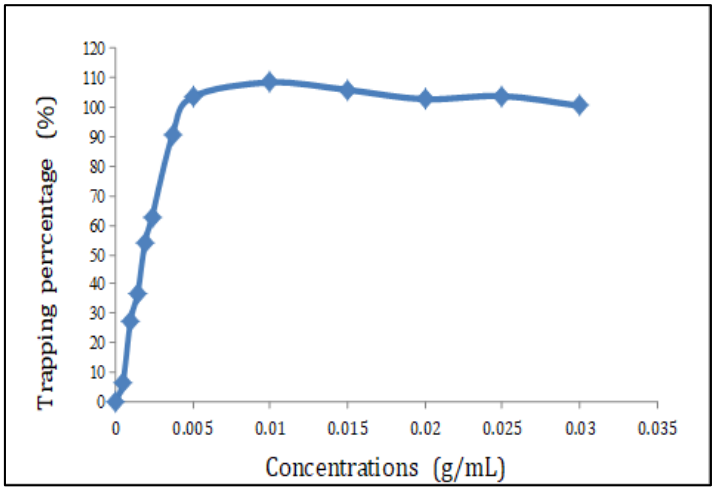

Figure 1 Percentage of DPPH radical scavenging by the ethanolic extract of *Detarium microcarpum* leaves

3.3.2. Annona senegalensis

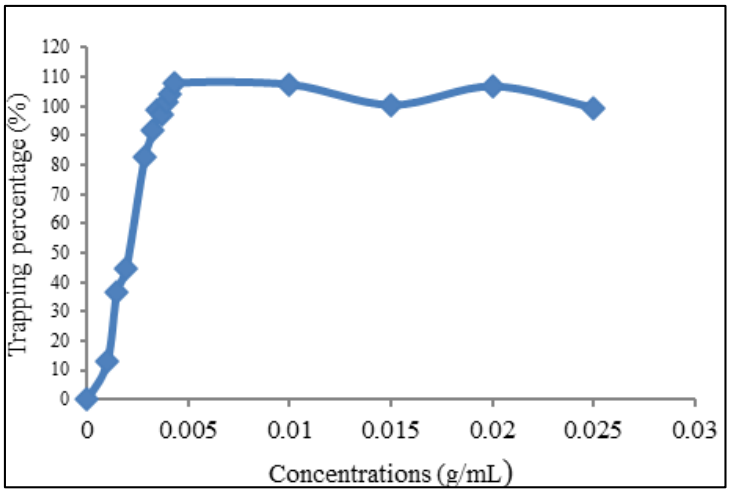

Figure 2 Percentage of DPPH radical scavenging by ethanolic extract of *Annona senegalensis* leaves

Figure 2 shows the DPPH radical scavenging percentage curve as a function of concentrations of hydroehanolic extract of *Annona senegalensis* leaves. A progressive increase in radical scavenging percentage was observed before becoming almost constant around 100%. The concentration of extract required to scavenge 50% of radical, determined from this curve, is 0.22 mg/mL. Similarly, the work of Winsou *et al*.[46] in Benin also showed that *Annona senegalensis* leaves have significant antiradical activity.

3.4. Total Inhibition Diameters of hydroethanolic Extract of *Detarium microcarpum* Leaves on Some Microbial Strains

Table 4 shows inhibition diameters of hydroethanolic extracts of *Detarium microcarpum* and *Annona senegalensis*, along with ciprofloxacin and amoxicillin, against 16 microbial strains. From this table, the hydroethanolic extract of *Annona senegalensis* leaves inhibited all clinical strains (12 strains) and reference strains (4 strains). Traoré *et al*.[15] demonstrated that hydroethanolic extract of *Annona senegalensis* leaves collected in Côte d'Ivoire inhibited *Streptococcus pneumoniae* strain. Our results align with previous studies, which showed that extracts of *Annona senegalensis* leaves inhibited various pathogens, including *Staphylococcus aureus*, *Candida albicans*, *Streptococcus mutans*, and *Aspergillus niger*, with significant inhibition zone diameters [47], [34]. These results corroborate previous studies that showed hydroethanolic extract of *Detarium microcarpum* inhibited microbial strains with significant inhibition zone diameters [48],[49]. The extracts of *Annona senegalensis* and *Detarium microcarpum* inhibited 100% of all tested microbial strains, whereas ciprofloxacin and amoxicillin inhibited 75% and 25% of targeted strains, respectively. It was observed that hydroethanolic extracts of *Detarium microcarpum* and *Annona senegalensis* have a broader spectrum of activity than the synthetic products used (ciprofloxacin and amoxicillin). The diversity of secondary metabolites in these plants could explain this remarkable activity. Similarly, inhibition of various microbial strains by hydroethanolic extracts of *Annona senegalensis* and *Detarium microcarpum* may justify use of these plants in treatment of urinary tract infections [49].

Table 4 Inhibition diameters (mm) of hydroethanolic plant extracts and tested antibiotics

	KP			***SA***				***EC***				***SD***			***CA***	***SP***
	VS	**S**	**UR**	**VS**	**S**	**UR**	**Re**	**VS**	**S**	**UR**	**Re**	**VS**	**S**	**UR**	**Re**	**Re**
Extract of D m	18.5±0.5	15±2	16±0	14.5±0.5	13±0	14±1	13.5±1,5	13.5±1,5	15±0	25.5±0,5	16±0	19.5±0,5	16.5±1,5	10±0	15±2	17±0
Extract of AS	21.5+1.5	19±1	18±1	18.5±0.5	17±1	22.5±2,5	16.5±0.5	18.5±0,5	16±1	18.5±0,5	20±0	7±1	21.5±1,5	13.5±1.5	15.5±0.5	14.5±2.5
A	19.5+0.5	16±1	-	-	-	-	-	13±0	-	-	-	-	13.5±1.5	-	-	-
C	41.5+0.5	38±0	36.5±0.5	18.5±0.5	18.5±1.5	38±0	-	41.5±3.5	43.5±0.5	14.5±0.5	38±0	-	40±0	40±0	-	-

Legends: Dm: Detarium microcarpum; AS: Annona senegalensis; VS: vaginal swab; S: sperm; UR: urine; Re: reference; SD: Streptococcus D; KP: Klebsiella pneumoniae; CA: Candida albicans; EC: Escherichia coli; SA: Staphylococcus aureus; SP: Streptococcus pneumoniae; A: amoxicillin; C: ciprofloxacin

3.5. Minimum Inhibitory Concentrations (MIC) and Minimum Bactericidal Concentrations (MBC) of extracts of two plants on tested strains

Table 5 presents the MICs and MBCs of hydroethanolic extract of *Detarium microcarpum* leaves, *Annona senegalensis*, ciprofloxacin, and amoxicillin. The MICs range from 0.39 mg/mL to 1.56 mg/mL, while MBCs fluctuate from 1.56 mg/mL to 12.5 mg/mL. Hydroethanolic extract of *Annona senegalensis* showed bactericidal activity on the 16 tested strains, including 12 clinical strains and 4 reference strains. Previous studies align with results obtained in this study[15]. However, our results differ from those of Yakubu *et al*.[34], who found higher MBCs for the hydroethanolic extract of *Annona senegalensis* leaves compared to our results with the *Staphylococcus aureus* strain. Regarding hydroethanolic

extract of *Detarium microcarpum* leaves, MICs range from 0.39 mg/mL to 1.56 mg/mL, with MBCs varying from 1.56 mg/mL to 12.25 mg/mL. Based on the obtained results, hydroethanolic extract of *Detarium microcarpum* leaves was found to be bactericidal against all tested strains. The findings of this study are in agreement with previous studies that demonstrated hydroethanolic extract of *Detarium microcarpum* leaves has a broad spectrum of antimicrobial activity [48], [49]. On the other hand, Fotso *et al*.[50] found higher minimum bactericidal concentrations with several strains compared to results of this study. Regarding antimicrobial activity of synthetic antibiotics (ciprofloxacin and amoxicillin), the MICs of ciprofloxacin range from 0.125 to 5 mg/mL, while MBCs range from 0.31 to 20 mg/mL. Amoxicillin inhibited only strains of *Klebsiella pneumoniae* (vaginal sample), *E. coli* (vaginal sample), *Streptococcus D* (sperm sample), and E. coli (ATCC 25922) at an inhibitory concentration of 10 mg/mL. However, it showed bactericidal activity against *Klebsiella pneumoniae* strain (sperm sample) at a concentration of 20 mg/mL. The results indicate that hydroethanolic extracts of *Detarium microcarpum* and *Annona senegalensis* leaves demonstrated more pronounced antibacterial activity than two synthetic antibiotics used (amoxicillin and ciprofloxacin). The diversity of secondary metabolites, particularly tannins, flavonoids, coumarins, anthraquinones, sterols, and triterpenes in the leaves of *Detarium microcarpum* and *Annona senegalensis*, could explain the significant antimicrobial activity observed [51],[52];[39],[41].

Table 5 Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC)

Strains	**Origin**	Extract of Dm		Extract of *AS*		**A**		**C**	
		Concentrations (mg/mL)							
		MIC	**MBC**	**MIC**	**MBC**	**MIC**	**MBC**	**MIC**	**MBC**
KP	VS	0.39	1.56	1.56	6.25	10.0	-	0.63	2.50
	S	1.56	5.00	0.39	1.56	5.00	20.00	0.63	2.50
	UR	0.78	5.00	1.56	6.25	-	-	1.25	5.00
SA	VS	0.78	3.13	0.78	3.13	-	-	0.16	0.63
	S	0.78	3.13	1.56	6.25	-	-	5.00	20.00
	UR	1.25	12.25	0.39	1.56	-	-	2.50	10.00
	ATCC 29213	0,39	1.56	0.39	6.25	-	-		
EC	VS	0.39	1.56	0.78	5.00	10.00	-	1.25	5.00
	S	0.39	1.56	0.39	6.25	-	-	0.16	0.31
	UR	0.78	6.25	0.39	12.50	-	-	0.16	0.63
	ATCC 25922	0.39	6.25	0.78	3.13	10.00	-	2.50	10.00
SD	VS	0.78	3.125	0.39	12.50	-	-	-	-
	S	0.78	3.125	0.78	3.13	10.00	-	1.25	5.00
	UR	0.78	3.125	0.39	10.00	-	-	2.50	10.00
CA	MHMR	0.39	1.56	10.00	-	-	-	-	-
SP	ATCC 4961	0.39	6.25	0.39	1.56	-	-	0.63	2.50

Legends: Dm: Detarium microcarpum; AS: Annona senegalensis; VS: vaginal swab; S: sperm; UR: urine; Re: reference; SD: Streptococcus D; KP: Klebsiella pneumoniae; CA: Candida albicans; EC: Escherichia coli; SA: Staphylococcus aureus; SP: Streptococcus pneumoniae; A: amoxicillin; C: ciprofloxacin

4. Conclusion

Natural substances extracted from plant biomass offer multiple benefits, which are utilized in food, cosmetics, and pharmaceutical industries. Infectious diseases are a serious health problem worldwide, particularly in Benin. Today, there is great interest in the use of medicinal plants to treat these infections. The present work aims to highlight two plants (*Annona senegalensis*, *Detarium microcarpum*) used in traditional medicine in Benin to treat urinary infections by determining their phytochemical potential and evaluating biological activities of their extract. According to results obtained, leaves of *Annona senegalensis* and *Detarium microcarpum* contain anthraquinones, catechic tannins,

coumarins, flavonoids, reducing compounds, sterols, and terpenes. Hydroethanolic extract of *Annona senegalensis* and *Detarium microcarpum* leaves showed bactericidal activity against sixteen (16) microbial strains used in this study. Ciprofloxacin and amoxicillin showed bactericidal activity against 75% and 25% of the tested strains, respectively. Hydroethanolic extract of *Annona senegalensis* and *Detarium microcarpum* exhibited more pronounced antimicrobial activity than two synthetic antibiotics used. The diversity of secondary metabolites, particularly tannins, flavonoids, coumarins, anthraquinones, sterols, and triterpenes present in the leaves of *Detarium microcarpum* and *Annona senegalensis*, could explain observed antimicrobial activity, which also supports the use of these plants in traditional medicine.

Compliance with ethical standards

Disclosure of conflict of interest

The authors declare that there is no conflict of interest regarding publication of this research paper.

References

[1] Dassou H, Ogni C, Yédomonhan H, Adomou A, Tossou M, Dougnon J, Akoègninou A. 2014. Diversity, veterinary uses and vulnerability of medicinal plants in North Benin. Int. J. Biol. Chem. Sci., 8(1): 189-210

[2] Jiofack T, Fokunang C, Guedje N, Kemeuze V, Fongnzossie E, Nkongmeneck BA, Ma-pongmetsem PM, Tsabang N. (2010). Ethnobotanical uses of medicinal plants of two ethnoecological regions of Cameroon. International Journal of Medicine and Medical Sciences, 2(3): 60- 79 ;

[3] Effoe Stéphane, Gbekley Efui Holaly, Mélila Mamatchi, Aban Amégninou, Tchacon-do Tchadjobo, Osseyi Elolo, Karou Damintoti Simplice et Kokou Kouami (2020). Ethnobotanical study of food plants used in traditional medicine in the Maritime region of Togo. Int. J. Biol. Chem. Sci. 14(8): 2837-2853;

[4] Magdalena Mititelu , Gabriel Olteanu, Sorinel Marius Neacs, Iuliana Stoicescu , Deni-sa-Elena Dumitrescu, Emma Gheorghe, Monica Tarcea, tefan Sebastian Busnatu , Corina-Bianca Ionit, Mîndrican , Ovidiu Tafuni, Ionela Belu , Antoanela Popescu , Sergiu Lupu and Carmen Elena Lupu (2024). Incidence of Urinary Infections and Be-havioral Risk Factors. Nutrients , 16, 446: 2-20;

[5] Sekhsokh Y., Chadli M., S.A. El Hamzaoui (2008). Frequency and sensitivity to antibiotics of bacteria isolated in urine. Medicine and infectious diseases. 38 324-327 ;

[6] Perry A, Morris M, Unsworth C, Duckett S, Skeat J, Dodd K, Taylor N, Reilly K: (2004) Therapy outcome measures for allied health practitioners in Australia: the AusTOMs. International Journal for Quality in Health Care, 16:1-7 ;

[7] Ouardi Raouia (2019). The current bacteriological profile of urinary infection and the status of antibiotic resistance. Cadi Ayyad University Faculty of Medicine and Pharmacy Marrakech. DOCTORATE IN MEDICINE

[8] Schoen EJ, Colby CJ, Ray GT (2000). Newborn circumcision decreases incidence and costs of urinary tract infections during the first year of life. Pediatrics.; 105(4 Pt 1):789-93;

[9] Onoh Linus UM (2021). Pattern of urinary tract infection in an all-female secondary school in Enugu metropolis, south east Nigeri. International Journal of Multidiscipli-nary Research and Growth Evaluation. 2(6); 145-147;

[10] Hantoosh Saad M. (2024). The Prevalence of Bacterial Urinary Tract Infections Among School-age Children. European Journal of Medical Genetics and Clinical Bio-logy, 1(5): 44-53;

[11] Petca, R.C.; Mares, , C.; Petca, A.; Negoit,ă, S.; Popescu, R.I.; Bot, , M.; Barabás, E.; Chibelean, C.B (2020). Spectrum and Antibiotic Resistance of Uropathogens in Ro-manian Females. Antibiotics 9, 472;

[12] Behzadi, P.; Behzadi, E.; Yazdanbod, H.; Aghapour, R.; Akbari Cheshmeh, M.; Sa-lehian Omran, D. A Survey on Urinary Tract Infections Associated with the Three Most Common Uropathogenic Bacteria. Maedica 2010, 5, 111–115;

[13] Zerbo P, Rasolodimby JM, Ouedraogo ON, Van Damme P. (2011). Plantes médicinales et pratiques médicales au Burkina Faso: cas des Sanan. Bois & Forets des Tropiques, 307 : 41-53 ;

[14] Kouyaté AM. (2005). Ethnobotanical aspects and study of the morphological and phenological variability of Detarium microcarpum Guill & Perr. (Mali). Doctoral thesis, University of Gent, p. 190;

[15] Traoré Y., Ouattara K., Yéo D., Doumbia I., Coulibaly A. (2019). Research into the antifungal and antibacterial activities of the leaves of Annona senegalensis Pers. (Annonaceae). J. Appl. Biosci 58, 4234–4242 ;

[16] Koudoro, Y. A., Alfred N. T., Selcuk K., Ozgur C., Agbangnan Dossa C. P., Avlessi F., Sohounhloue C. K. D., Mehmet E. Duru, Rodica M. D. (2022). Phenolic profiles, antioxidant, antiquorum sensing, antibiofilm and enzyme inhibitory activities of selected Acacia species collected from Benin. LWT- Food Science and technology/Elsevier. LWT171, 114162;

[17] Koudoro Yaya Alain, Daye Efloric Raphaël, Dassou Hospice Gbèwonmèdéa, Atindehou Ménonvè, Ag-bangnan Dossa Cokou Pascal, Alitonou Guy Alain, Félicien Avlessi, Dinica Rodica Mihaela, So-hounhloué C. K Dominique (2021). Phytochemical screening, antioxidant capacity, antibacterial and anti-inflammatory activities of ethanolic extract of *Cordia senegalensis* leaves, a plant used in Benin to treat skin diseases. Chemistry Research Journal, 6(2):137-146;

[18] Dohou, N., Yamni, K., Tahrouch, S., Hassani, L. M. I., Bodoc A. & Gmira, N., (2003). Phytochemical screening of an endemic Ibero-Moroccan, Thymelaea lytroides''Bull. Soc. Pharm. Bordeaux, 142, 61- 78 ;

[19] Agbangnan D.C.P., Tachon C.B., Chrostowka A., Fouquet E., Sohounhloue D.C.K.. (2012).''Phytochemical study of a tinctorial plant of benin traditional pharmacopoeia: The red sorghum (*sorghum caudatum*) of Benin'', Scientific Study & Research, 13(2), pp.121-135;

[20] Lupoae P., Cristea1 V., Borda D., Lupoae M., Gurau G. and Dinica R. M. (2015). Phytochemical Screening: Antioxidant and Antibacterial Properties of Potamogeton Species in Order to Obtain Valuable Feed Additives. Journal of Oleo Science, pp : 1-13;

[21] Singleton, V.L., Orthofer, R., Lamuela-Raventos, R.M. (1999): Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. Methods Enzymol. 299, 152-178 ;

[22] Mbacke D.I.S, FalL A.D, Diatta-Badji K., Sarr A, Madieye S., Moussa S., Mbaye A, Diatta W et Bassene E. (2017). Evaluation of the antioxidant activity of hydro-ethanolic extracts of leaves and bark of Piliostigma thonningii Schumach. Int. J. Biol. Chem. Sci. 11(2): 768-776 ;

[23] Djeridane, A., Yous, M., Nadjemi, B., Boutassouna, D., Stocker, P., Vidal, N. (2006). Antioxidant activity of some Algerian medicinal plants extracts containing phenolic compounds. Food Chem. 97: 654-660 ;

[24] Makkar H.P.S. (2000). Recent advances in the in vitro gas method for evaluation of nutritional quality of feed resources. Animal Production and Health Section Joint FAO/IAEA Division International Ato-mic Energy Agency Vienna (Austria) ;

[25] Chabi-Sika K., Sina, H., Boya, B., Bade, F., Hounnou, T., Badoussi, M. E., Adjatin A., Baba-Moussa L (2021). Richardia brasilensis collected in Southern-Benin: Phytochemical, Antimicrobial Activity and Toxicity. Asian Journal of Biology, 13(4):22-23;

[26] Amoussa, A. M. O., Sanni, A., Lagnika L. (2015). Antioxidant activity and the estimation of total phenolic, flavonoid and flavonol contents of the bark extracts of Acacia ataxacantha. Journal of Pharmacognosy and Phytochemistry, 4(2): 172-178 ;

[27] Loubaki B. C., Ouattara A. S., Ouattara A. T., Ouedraogo R., Traore A. S. (2012) Rev. CAMES - Série, 66 -73;

[28] Saxena M., Saxena J., Nema R., Singh D. & Gupta A., (2013) Journal of Pharma-cognosy and Phytochemistry, 1 (6) 168 -182;

[29] Ebi G. C., and Afieroho O. E. (2011) Phytochemical and antimicrobial studies on *Detarium microcarpum* African Journal of Biotechnology, 10(3) 457 -462;

[30] kabassi B. S., Jospin A. D., Tchobo P. F., Tchatcha D. A., Houénon G. H. A. , Yovo M., Dédjiho C. C., Bogninou-Agbidinoukoun R. S. G., Soumanou M. M. (2021). Phytochemical screening and evaluation of the antiradical and antimicrobial activities of the organs of Detarium microcarpum Guill. & Perr from the Sudanian zone of Benin. J. Soc. Ouest-Afr. Chim. 050 ; 68- 75;

[31] Hamadou H. H, Moussa I, Ikhiri K,.Ouedraogo B, Adamou R. (2018). Phytochemical screening and determination of polyphenols from Detarium microcarpum Guill. and Perr. used in the treatment of parasitic diseases in Niger 14 (5), 390-399;

[32] Dembele K. T., Malviya N. and Mahajan S. (2013). Preliminary Phytochemical Screening of Bark of Some Important Trees of College Campus With Special Reference to Tannins, Glycoside and their Medicinal Properties, International Research Journal of Environment Science. 2 (11) : 13-16;

[33] Abdullahi, A. R., Malami, S., & Bichi, L. A. (2021). In vivo antiplasmodial activity of *Detarium microcarpum* (fabaceae) stem bark extract. Thrita, 10(1), 1-9;

[34] Yakubu M., Yahi D., Mbaya Y. P., Bargu J. S., Dibila H. M., Igwenagu E.N., Ba-dau S. J., Ndahi J. J., Auwal M. S. and Dibila H. M. (2021) Acute toxicity, chemi-cal compositions and effect of aqueous extract of Moringa oleifera stem bark on some haematological parameters. IJEHMR4:716-732;

[35] Diallo F. (2020). Phytochemical study and anti-radical activity of three medicinal plants used in the treatment of uterine fibroids in Mali. Doctoral thesis at the Faculty of Pharmacy of Bamako; p 101;

[36] Mbaya, Y. P., Bobbo, A. G., Barkindo, A. A. and Hayatu, A. I. (2023) Toxicity and the Effects of Ethanolic Leaf Extract of Annona senegalensis on Haematological Parameters in Albino Rats;

[37] Manolaraki F. (2011). Anthelmintic properties of sainfoin (Onobrychisviciifoliae). Analysis of the variation factors and the role of the phenolic compounds involved. Doctoral thesis from the University of Toulouse III, Toulouse;

[38] Badiaga M. (2011). Ethnobotanical study, phytochemistry and biological activities of Nauclea latifolia smith, an African medicinal plant collected in Mali. Doctoral thesis, University of Bamako. 183;

[39] Duong Q. P., Hieu T. P., Jae W. H., Tung H. N., Huong T. N., Thi D. N., Thu T. T. N., Cuong T. H., Hong M. P., Hoang D. V., Gyung J. C., Quang L. D. (2021) Extracts and metabolites derived from the leaves of Cassia alata L. exhibit in vitro and in vivo antimicrobial activities against fungal and bacterial plant pathogens. Industrial Crops & Products 166, 113465;

[40] Fikri H., Fechtali T., Timinouni M., Zouheir Y. and Mamoumi M. (2022) Struc-ture activity modeling of essential oils compounds and plant secondary metabolites: a Mini review of Antimicrobial Activity. Arabian Journal of Medicinal & Aromatic Plant, 85-95 ;

[41] Mohammed A. E, Sahar S. A., Ashwag S., Rasha S. S., Kawther A., Modhi O. A., Ishrat R. (2023). In silico Prediction of Malvaviscus arboreus Metabolites and Green Synthesis of Silver Nanoparticles – Opportunities for Safer Anti-Bacterial and Anti-Cancer Precision Medicine. International Journal of Nanomedicine, 18, 2141-2162;

[42] Lawaly M. M., Obadoni, B. O. and Ochuko, P. O. (2001). Phytochemical studies and comparative efficacy of the crude extracts of some homeostatic plants in Edo and Delta States of Nigeria. Global. J. Pure Applied Sciences. 8: 203-208;

[43] David E., Afolabi P., Olotu j., Ojerinde S., Agwom F., and Ajima U. (2017). Phytochemical analysis, antidiabetic and toxicity studies of the methanolic leaf extract of Detarium micro-carpum guill and perr in wistar albino rats, Journal of Chemical and Pharmaceutical Research, 9(11) ;

[44] Daddona P.E., wright J.L., Hutchinson C.R. (1976). ''Alkaloid catabolism and mobilization in Catharanthusroseus'', Phytochem, 15 : 941-945;

[45] Manolaraki F. (2011). 'Anthelmintic properties of sainfoin (Onobrychisviciifoliae). Analysis of the variation factors and the role of the phenolic compounds involved. Doctoral thesis from the University of Toulouse III, Toulouse

[46] Winsou Irène, Anago Eugenie, Dah-Nouvlessounon Durand, Aclinou Eustache, Nounagnon Martial, Roko Gautier, Baba-Moussa Lamine (2022). Phenolic Profile and Antioxidant, Anti-Inflammatory Activity of Annona senegalensis, Ipomoea batatas, Terminalia superba and Psidium guajava Linn Extracts Used in Benin. American Journal of Plant Sciences 13 (9) 1296-1310;

[47] Abubakar M. S, Balogun E., Abdurahman E. M., Niok A. J., Shok M., Moham-mad A.,Garba M. (2006). Ethnomedical treatment of Poisonous Snakebites: Plant ex-tracts Neutralized Naja nigricollis Venom. Pharmaceutical Biology, 44(5). 343-348;

[48] Dahiru M., Kolawole S., Tukur A. (2022). Antibacterial and Phytochemicals Status of *Detarium microcarpum* (Guill and Perr) Stem Bark. Natural Resources for Human Health 4:1-6. https://doi.org/10.53365/nrfhh/145597;

[49] Sanusi S. M., Lawaly M., Usman A., Musa F. M., Ardo B. (2022). Phytochemical Analysis and Antibacterial Activity of Stem Bark Extracts of *Detarium microcarpum* Against Bacteria Causing Gastrointestinal Tract Infections in Humans Dutse Journal of Pure and Applied Sciences (DUJOPAS);

[50] Fotso J. A. Y., Takuissu G. R. N., Ngondi J. L., Oben J. E. (2021). Acute and subchronic toxicity studies of aqueous bark extract of *Detarium microcarpum* guill. and perr in albinos wistar rat. Toxicology and Environmental Health Sciences /10.1007/s13530-022-0013,9-4 ;

[51] Shan B., Yi-Zhong C. , Brooks J. D., Corke H. (2007). The in vitro antibacterial activity of dietary spice and medicinal herb extracts, nternational Journal of Food Mi-crobiology 117 :112 – 119.

[52] Vincken J. P., Heng L., De Groot A., Gruppen H. (2007). Review saponins, classi-fication and occurrence in the plant kindom. Phytochemistry, 68, Pp: 275-297.