Asian Journal of Applied Chemistry Research

Volume 14, Issue 1, Page 35-43, 2023; Article no.AJACR.102805
ISSN: 2582-0273

Phytochemical, *in vitro* Antimicrobial, Proximate, Anti-inflammatory, Antioxidant and Anti-hyperglycemic Activities of Root Extracts of *Combretum platypterum* (Welw)

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/AJACR/2023/v14i1257

Open Peer Review History:
This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/102805

Original Research Article

Received: 18/05/2023
Accepted: 07/07/2023
Published: 20/07/2023

ABSTRACT

Aim: This work was to determine the phytochemical, proximate, *in vitro* antimicrobial, antioxidant, anti-inflammatory, and anti-hyperglycemic activities of root extracts of *Combretum platypterum.*
Methods: Roots of *C. platypterum* were cold extracted with methanol, ethyl acetate, and n-hexane. The extracts were subjected to proximate, phytochemical analyses, antimicrobial, antioxidant, anti-inflammatory, and anti-hyperglycemic assays.
Results: The result of proximate analysis revealed that the roots contained 73.90±0.10% carbohydrates, the mean % of fibre and ash contents were >7.00, whereas the average protein and

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moisture contents were >4.00. The result of qualitative phytochemical analysis revealed that alkaloids, saponins, tannins, phenolic compounds and carbohydrates were present in the three extracts, whereas steroids were sparingly present only in n-hexane extract. The result of antimicrobial screening indicated that *B. subtilis*, *S. aureus, E. coli, C. albicans* and *S. typhi* were susceptible to the inhibitions of the three extracts in concentration-dependent manner, whereas only the n-hexane extract showed a dose-dependent inhibition against *P. aeruginosa***.** Methanol and ethyl acetate extracts showed good anti-inflammatory as well as antioxidant activity. The inhibition of α-amylase was greater with n-hexane extract.

Conclusion: the extracts showed good antimicrobial, anti-inflammatory, antioxidant and anti-hyperglycemic activities and can be a potential antimicrobial, anti-inflammatory, antioxidant and anti-hyperglycemic agent.

Keywords: Combretum platypterum; anti-hyperglycemic; anti-inflammatory; antioxidant; antimicrobial; phytochemical; proximate.

1. INTRODUCTION

From the time immemorial, medicinal plants have been used in treating various types of diseases, and through scientific researches to verify their efficacy, plant-based medicines have been produced [1]. In the beginning, the trial and error method was used to treat illnesses or even simply to feel better, and in this way, to distinguish useful plants with beneficial effects [2].

Combretum platypterum has been used in Nigeria traditional medicine in treating various forms of diseases such as helminthiasis, sexually transmitted diseases, conjunctivitis, malaria, lumps, fever, eye problems, diarrhea, lower backache, coughs and swellings [3].

Despite the ethnomedicinal uses of *C. platypterum,* not much scientific works have been done on the plant to validate the traditional uses.

2. MATERIALS AND METHODS

Collection and Identification of the Plant: The roots of *Combretum platypterum* were collected within the surroundings of the Adada River in Nsukka Local Government Area, Enugu State, Nigeria. The collected plant roots were identified and authenticated by Mr. Alfred Ozioko (The Chief Taxonomist) at the International Center for Ethno medicine and Drug Development. Herbarium specimens were deposited in the herbarium of the International Center for Ethno medicine and Drug Development (Voucher number: Intercedd/260510).

Preparation of Plant Extract: The extraction was carried out using cold extraction method. The *Combretum platypterum* roots were washed, air-dried at room temperature and ground into powder. 1 kg of dry root powder was then macerated with 6 liters, each of methanol, ethyl acetate and n-hexane for 48 hours in an air-tight container at room temperature. The mixtures were filtered with a glass funnel embedded with cotton wool into a beaker and evaporated to dryness using a rotary evaporator at 40 °C and labeled CpRM (methanol extract), CpRE (ethyl acetate extract), and CpRH (n-hexane extract). The extracts obtained were kept at 4 ^{0}C until further use.

Phytochemical Analysis: Freshly prepared extracts were subjected to quantitative and qualitative phytochemical analysis to determine the presence or absence of flavonoids, alkaloids, terpenoids, saponins, carbohydrates, resins, tannins, reducing sugars, glycosides, and proteins according to the methods described by [4-8] with slight modifications.

Proximate Analysis: Proximate analysis of the powdered root for moisture, ash, fiber, protein, oil, and carbohydrate content was determined using standard methods AOAC (2010) [9] as described by [10].

2.1 Antimicrobial Assay

Samples: The antimicrobial sensitivity of the three samples from the plant extracts was determined using the agar-well diffusion method [11-12].

Test Organisms: The test organisms were obtained from the Department of Pharmaceutical Microbiology and Biotechnology, Faculty of Pharmaceutical Sciences, Nnamdi Azikiwe University, Awka, Nigeria. Bacteria strains of

Escherichia coli, *Staphylococcus aureus*, *Klebsiella pneumonia, Streptococcus pneumonia, Candida albicans*, and *Aspergillus niger* were used in the study.

Culture Media and Other Reagents Used in Microbiological Analyses: Nutrient Broth, Nutrient agar, Sabouraud Dextrose Agar, Mueller Hinton Agar and Sabouraud Dextrose Broth (Oxoid Limited, England) were the culture media used. The culture media were prepared according to the direction of the manufacturers.

2.2 *In vitro* Antioxidant Assay

DPPH Radical Scavenging Assay was determined using DPPH as previously described by Shimada et al. [13]. The FRAP test was performed according to the method described by Benzie and Strain [14], whereas, the ABTS radical scavenging assay was conducted in accordance with the method outlined by Re et al. [15] with a minor modification made by Siddhuraju et al. [16]. Nitric oxide radical scavenging potential of the plant extracts were examined according to the procedure described by Marcocci et al. [17].

2.3 *In vitro* Anti-Inflammatory Screening

Heat-Induced Hemolysis screening was carried out using the method as described by Okoli et al. [18]. Protein denaturation assay of the CpRH, CpRE and CpRM was done according to the procedure described by Banerjeee et al. [19], whereas the proteinase inhibitory assay was done using the method described by Dabhade et al. [20]. The root extracts of *Combretum platypterum* were tested for their ability to inhibit lipoxygenase using the technique described by Anosike et al. [21].

2.4 *In vitro* Antihyperglycemic Screening

Alpha-amylase inhibitory and yeast cell uptake of glucose screenings of the root extracts of *Combretum platypterum* was done according to a method previously described by Anarado et al. [3], Ranila et al. [22]

3. RESULTS AND DISCUSSION

3.1 Proximate Composition of *C. platypterum* Whole Root Sample

The result for the proximate composition of *C. platypterum* whole root sample was presented in Table 1.

The result whole root sample of *C. platypterum* (Table 1) showed that carbohydrates (73.90 %) and crude fibre (7.6333%) were the highest concentrations in *C. platypterum* root, followed by ash (7.55 %), moisture (4.33%), proteins (4.20%) while oil was the least content (2.26%).

3.2 Qualitative and Quantitative Phytochemical Composition of Whole Root extracts of *C. platypterum*

The results of preliminary qualitative and quantitative phytochemical analyses of whole root extracts of *C. platypterum* were shown in Tables 2 and 3.

The result of qualitative phytochemical screening of *C. platypterum* root extracts (Table 2) showed that alkaloids, saponins, carbohydrates, phenolic compounds and tannins were present in all the extracts. Steroids were sparingly present only in n-hexane extract, which was not surprising since “like dissolves like”- hydrophobic steroids would only be soluble in non- polar solvent such as n-hexane. The high abundance of flavonoids only in n-hexane extract showed an absence of hydrophilic flavonoid glycosides such as quercetin [23]. The solubility of the flavonoids in the n-hexane extracts was also in line with the report of Chaves et al. [24]. that less hydrophilic flavonoids such as aglycones of isoflavones, flavanones, methylated flavones could be extracted with n-hexane. Tannins were found more in ethyl acetate and n-hexane extracts, and

Table 1. Proximate composition of *C. platypterum* whole root sample

Parameter	Root Composition (%)
Moisture	4.33± 0.11
Ash	7.55± 0.16
Protein	4.20± 0.40
Fiber	7.63± 0.35
Oil	2.26± 0.05
Carbohydrate	73.90± 0.10

Table 2. Qualitative phytochemical composition of whole root extracts of *C. platypterum*

Phytochemical Constituents	CpRH	CpRE	CpRM
Alkaloids	+	++	++
Saponins	+++	++	+++
Tannins	++	+++	+
Flavonoids	+++	-	-
Steroids	+	-	-
Terpenoids	-	++	-
Cardiac Glycosides	-	+	+
Carbohydrates	+	+	+
Resins	-	-	-
Phenolics	+	+++	+++
Reducing Sugars	-	+	-
Proteins	-	-	-
Anthocyanins	-	+++	+++

Table 3. Results of quantitative analyses of whole root sample of *C. platypterum*

Phytochemical Constituents	Root Quantity (mg/100g)
Alkaloids	8.82±0.30
Flavonoids	1.06±0.08
Saponins	34.17±0.29
Tannins	18.37±0.55
Phenolics	100.53±3.58
Steroids	0.13±0.02
Terpenoids	3.30±0.03

slightly in methanol which was surprising, considering that tannins which are polyphenolic compounds were expected to be most in polar extracts. The polarity of the solvents also played role in the extraction of polyphenolic compounds and anthocyanins [25]. The amphiphilic nature of saponins was also observed in their high abundance in both methanol and n-hexane extracts [26]. Proteins, oils and resins were absent in all the extracts.

The result of quantitative phytochemical analysis of *C. platypterum* root sample (Table 3) showed that phenolics, saponins and tannins were found in high abundance, while flavonoids were least in abundance.

3.3 Results of *in vitro* Antimicrobial Activities of *C. platypterum* Root Extracts

The results of antibacterial, antifungal and MIC analyses were shown in Tables 4, 5 and 6.

The result of antibacterial analysis of root extracts of *C. platypterum* (4) showed that ethylacetate extract inhibited the growth of *B. subtilis* in a dose-dependent manner. At 5 mg/mL and 2.5 mg/mL, the inhibition of the extract against *B. subtilis* and *S. aureus* was higher than the inhibition of ciprofloxacin at 50 µg/mL, while the methanol and n-hexane extracts showed mild activity against both organisms especially at higher concentrations. At 5 mg/mL, n-hexane and ethylacetate extracts inhibited the growth of *E. coli* more than ciprofloxacin at 50 µg/mL.

K. pneumonia was resistant to the activities of all the extracts in all concentration, while *P. aeruginosa* and *S. typhi* were mildly susceptible to the inhibition of the extracts in a concentration-dependent manner. The inhibition of the *S. aureus* by the methanol extract was against the report of Ogbole et al. [27], who reported that strains of methiciline-resistant *S. aureus* were resistant to methanol extract of the plant.

The result of antifungal analysis of *C. platypterum* root extract (5) showed that *A. niger* was only mildly susceptible to activity of methanol extract of the plant at 5 mg/mL, and was resistant to the inhibitions of other extracts in all concentrations. The resistance of *A. niger* to many concentrations of methanol extract was in line with what was obtained with the methanol leaf extract of the same plant. Ethyl acetate

Table 4. Result of antibacterial screening of *C. platypterum* whole root extracts (mm)

Extract	*B. subtilis*	*K. pneumonia*	*P. aeruginosa*	*S. aureus*	*E. coli*	*S. typhi*
CpRM-5.0 mg/mL	12.17±0.47	0.00±0.00	7.00±0.00	22.2±0.87	10.20±0.26	17.00±0.00
CpRM-2.5 mg/mL	10.00±0.00	0.00±0.00	5.53±0.06	20.00±0.00	6.1±0.17	15.00±0.00
CpRM-1.0 mg/mL	7.00±0.00	0.00±0.00	4.03±0.06	13.00±0.00	3.00±0.00	10.00±0.00
CpRM-0.6 mg/mL	0.00±0.00	0.00±0.00	0.00±0.00	9.53±0.50	0.00±0.00	0.00±0.00
CpRH-5.0 mg/mL	22.40±1.04	0.00±0.00	33.33±1.53	15.33±0.58	22.00±0.00	10.00±0.00
CpRH-2.5 mg/mL	15.73±0.70	0.00±0.00	30.00±0.00	10.00±0.00	13.27±0.23	5.03±0.25
CpRH-1.0 mg/mL	0.00±0.00	0.00±0.00	26.9±0.173	0.00±0.00	6.50±0.50	0.00±0.00
CpRH-0.6 mg/mL	0.00±0.00	0.00±0.00	20.00±0.00	0.00±0.00	3.37±0.40	0.00±0.00
CpRE-5.0 mg/mL	47.7±1.14	0.00±0.00	0.00±0.00	34.67±1.53	26.33±1.16	16.00±0.00
CpRE-2.5 mg/mL	41.00±0.00	0.00±0.00	0.00±0.00	30.00±0.00	19.23±0.21	7.00±0.00
CpRE-1.0 mg/mL	34.33±1.53	0.00±0.00	0.00±0.00	20.33±0.58	10.97±1.00	4.10±0.17
CpRE-0.6 mg/mL	16.07±0.81	0.00±0.00	0.00±0.00	13.4±0.3605	7.54±0.227	0.00±0.00
Ciprofloxacin 50 µg/mL	38.93±1.00	41.67±2.0816	41.67±1.15	22.33±0.57	19.16± 0.28	35.57±0.30

and n-hexane extracts showed no activity against *A. niger* in all concentrations. *C. albicans* was sensitive to the activity of the extracts, with n-hexane extract showing highest activity (32.67±2.08 mm) at 5 mg/mL, higher than the activity of fluconazole (32.67±0.58 mm) at 50 µg/mL. The high activity of the n-hexane extract of the root against *C. albicans* was against the non-activity result obtained with the n-hexane leaf extract of the same plant.

Table 5. Result of *in vitro* antifungal activities of *C. platypterum* whole root extracts (mm)

Extract	*A. niger*	*C. albicans*
CPRM-5.0 mg/mL	5.70±0.26	15.87±0.46
CPRM-2.5 mg/mL	0.00±0.00	12.00±0.00
CPRM-1.0 mg/mL	0.00±0.00	10.00±0.00
CPRM-0.6 mg/mL	0.00±0.00	8.83±1.04
CPRH-5.0 mg/mL	0.00±0.00	32.67±2.08
CPRH-2.5 mg/mL	0.00±0.00	27.67±0.58
CPRH-1.0 mg/mL	0.00±0.00	21.33±0.58
CPRH-0.6 mg/mL	0.00±0.00	19.07±0.81
CPRE-5.0 mg/mL	0.00±0.00	26.33±1.15
CPRE-2.5 mg/mL	0.00±0.00	19.23±0.21
CPRE-1.0 mg/mL	0.00±0.00	10.97±1.00
CPRE-0.6 mg/mL	0.00±0.00	7.54±0.23
Fluconazole 50 µg/mL	12.00±0.00	32.27±1.36

Table 6. Result of minimum inhibitory concentration (mg/mL) of *C. platypterum* whole root extracts

Microorganism	CpRM	CpRE	CpRH	Control Drug
S. typhi	1.20	0.80	1.00	0.10
E. coli	1.00	0.10	0.10	0.50
S. aureus	0.50	0.50	2.00	0.10
P. aeruginosa	1.50	10.00	0.50	0.10
S. pneumonea	-	-	-	0.50
B. subtilis	2.50	0.50	0.50	0.50
C. albicans	0.50	0.50	0.50	0.10

3.4 *In vitro* Anti-Inflammatory Activities of *C. platypterum* Root Extracts

Table 7. Results of *in vitro* anti-inflammatory activities of *C. platypterum* root extracts

Sample concentrations	Heat-Induced Hemolysis(%)	Effect on Protein Denaturation(%)	Proteinase Inhibitory Activity(%)	Lipoxygenase Inhibition Assay(%)
CpRM-250 mg/mL	3.20±0.26	12.27±0.25	24.00±0.0	3.63±0.12
CpRM-500 mg/mL	12.13±0.15	28.33±1.53	17.00±0.00	8.33±0.21
CpRM-1000 mg/mL	30.00±0.00	35.03±0.06	29.40±0.10	12.20±0.17
CpRE-250mg/ml	0.93±0.12	3.33±0.40	2.33±0.29	14.30±0.20
CpRE-500 mg/mL	2.01±0.53	22.50±0.78	11.77±0.40	20.40±0.10
CpRE1000 mg/mL	6.23±0.25	25.17±0.29	26.30±1.56	30.31±0.60
CpRH-250 mg/mL	3.29±0.26	1.157±0.16	2.16±0.30	5.17±0.29
CpRH-500 mg/mL	8.47±0.47	7.60±0.69	7.00±0.00	16.20±0.26
CpRH1000 mg/mL	14.10±0.78	15.33±0.06	20.3±0.2645	27.47±0.90
Diclofenac -250 mg/mL	24.93±0.50	75.88±1.53	35.33±1.1547	45.00±0.00
Diclofenac -500 mg/mL	49.57±0.75	84.37±0.55	63.50±0.71	75.67±1.15
Diclofenac -1000 mg/mL	75±0	88.40±1.55	85.40±0.66	86.33±2.31

The result of anti-inflammatory screening of the root extracts of *C. platypterum* (Table 7) indicated that methanol extract showed anti-inflammatory activity through heat-induced hemolysis, protein denaturation and proteinase inhibition in concentration-dependent manner. At 100 mg/mL, methanol extract showed greater percentage heat-induced hemolysis than diclofenac at 250 mg/mL. The anti-inflammatory activity of the methanolic extracts could be as a result of the presence of phenolic compounds found in the extract [28]. The ethylacetate extract also inhibited lipoxygenase as well as proteinase in a dose-dependent manner. Lipoxygenase was inhibited by the hexane extract in concentration-dependent manner. The use of the plant in the treatment of inflammation is justified.

3.5 Result of *in vitro* Antioxidant Studies of *C. platypterum* Root Extracts

The result of antioxidant screening (Table 8) showed that the three extracts showed good antioxidant activity through ferric reducing antioxidant power at the highest concentration (1000 mg/mL). The activity of the n-hexane through nitric oxide was comparable to the activity of ascorbic acid which was used as the standard drug.

3.6 Result of *in vitro* Antihyperglycemic Assay of *C. platypterum* Root Extracts

The result of antidiabetic activity (Table 9) showed that methanol and ethyl acetate extracts

Table 8. Results of *in vitro* antioxidant activities of *C. platypterum* root extracts

Sample concentrations	Nitric Oxide (%)	DPPH (µg/mL)	FRAP (mgAA/g)	ABTS ASSAY (µmol/EAA/g)
CPRM-1000 mg/mL	0.15±0.00	1.64±0.12	10.68±0.10	1.16±0.10
CPRM-500 mg/mL	0.11±0.00	1.41±0.015	9.74±0.488	0.84±0.03605
CPRM-250 mg/mL	0.083±0.002	1.00±0.04	7.58±0.07	0.42±0.025
CPRE-1000 mg/mL	0.87±0.02	3.10±0.24	15.96±0.04	0.24±0.06
CPRE-500 mg/mL	0.55±0.00	2.06±0.10	9.79±0.05	0.15±0.00
CPRE250 mg/mL	0.18±0.01	1.00±0.00	9.00±0.00	0.00±0.00
CPRH-1000 mg/mL	1.97±0.00	4.40±0.15	23.50±0.7975	1.89±0.12
CPRH-500 mg/mL	1.36±0.02	3.30±0.14	20.06±0.10	1.05±0.06
CPRH250 mg/mL	1.00±0.00	2.95±0.09	19.27±0.25	0.00±0.00
Ascorbic acid-1000 mg/mL	5.37±0.15	45.97±0.96	85.88±0.29	16.20±0.18
Ascorbic acid-500 mg/mL	3.46±0.04	40.07±0.12	80.55±0.03	9.13±0.05
Ascorbic acid-250 mg/mL	2.14±0.16	37.43±0.1	68.15±0.26	5.03±0.06

Table 9. Results of *in vitro* anti-hyperglycemic activities of *C. platypterum* root extracts

sample concentrations	αAmylase inhibitory activities (%)	Glucose uptake (%)
CPRM-1000 mg/mL	23.38±0.78	28.93±0.05
CPRM-500 mg/mL	18.40±0.26	22.27±0.26
CPRM-250 mg/mL	8.26±0.28	17.64±0.16
CPRE-1000 mg/ml	27.54±0.24	31.26±0.96
CPRE-500 mg/mL	23.49±0.43	19.82±0.03
CPRE250 mg/mL	19.87±0.35	13.88±0.30
CPRH-1000 mg/mL	12.67±0.66	8.46±0.42
CPRH-500 mg/mL	7.073±0.34	4.45±0.03
CPRH250 mg/ml	2.39±0.18	1.71±0.45
Metformin -1000 mg/mL	53.27±0.46	68.00±0.00
Metformin -500 mg/mL	47.53±0.90	43.83±1.04
Metformin -250 mg/mL	40.40±0.66	27.00±2.00

showed good glucose uptake and also inhibit α-amylase in a dose-dependent manner. At 1000 mg/mL, both extracts were uptake glucose more than the standard drug- metformin at 250 mg/mL with uptake percentage of 28.93±0.05 and 31.26±0.96 respectively. The n-hexane extract only exhibited moderate activity.

4. CONCLUSION

Methanol, ethyl acetate and n-hexane extracts showed very good antimicrobial, antioxidant, anti-inflammatory and antidiabetic activities, which may be as a result of the metabolites present in them. Methanol extract showed best activity among the three extracts. Thus, the plant whole root can act as anti-inflammatory, antioxidant and antidiabetic agent.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Sofowora A, Ogunbodede E, Onayade A. The role and place of medicinal plants in the strategies for disease prevention. African journal of Traditional, Complementary, and Alternative Medicines : AJTCAM. 2013;10(5): 210–229. Available:https://doi.org/10.4314/ajtcam.v10i5.2
2. Salmerón-Manzano E, Garrido-Cardenas JA, Manzano-Agugliaro F. Worldwide research trends on medicinal plants. International Journal of Environmental Research and Public Health. 2020;17(10): 3376. Available:https://doi.org/10.3390/ijerph17103376
3. Anarado CE, Ajiwe VIE, Obumselu OF, Anarado CJO, Okafor NP. Phytochemical, Proximate, *in-vitro* Anti-malarial, Anti-inflammatory and Antimicrobial Screening of Leaf Extracts of *Combretum platypterum* (Welw) Hutch and Dalziel. Journal of Pharmaceutical Research International. 2021;33(52A): 39-61. DOI: 10.9734/JPRI/2021
4. Raaman N. Phytochemical techniques. New India Publishing Agency, New Delhi. 2006:19-22.
5. Ukoha PO, Cemaluk EAC, Nnamdi OL, Madus EP. Tannins and other phytochemicals of the Samanaeasaman pods and their antimicrobial activities. African Journal of Pure and Applied Chemistry. 2011;5(8):237-244.
6. Senguttuvan J, Paulsamy S, Karthika K. Phytochemical analysis and evaluation of leaf and root parts of the medicinal herb, *Hypochaerisradicata* L. for *in vitro* antioxidant activities. Asian Pacific Journal of Tropical Biomedicine. 2014;4(1):359-S367.
7. Sankhalkar S, Vernekar V. Quantitative and qualitative analysis of phenolic and flavonoid content in *Moringa oleifera* Lam and *Ocimum tenuiflorum* L. Pharmacognosy Research. 2016: 8(1):16 21.
8. Anarado CE, Anarado CJO, Umedum NL, Ogbodo QM. Comparative phytochemical and anti-microbial studies of leaf, stem, root of *Spathodea campanulata*. Asian Journal of Applied Chemistry Research. 2020;6(1):10-20. DOI: 10.9734/AJACR/2020/v6i130148
9. Anarado CE, Ajiwe VIE, Anarado CJO, Obumselu OF, Onuegbu TU, Okafor SE. Phytochemical, Proximate, In-vitro Antimalarial and Antimicrobial screening of leaf Extracts of Justicia carneaLindl. International Research Journal of Pure & Applied Chemistry. 2021;22(8): 55-73. DOI: 10.9734/IRJPAC/2021/v22i830428
10. Association of official analytical chemists (AOAC). Official methods of Analysis. Association of official chemists. 15th Edn. Association of Official Analytical Chemists, Washington DC. 2010;10-30.
11. Srinivasan D, Nathan S, Suresh T, Perumalsamy LP. Antimicrobial activity of certain Indian medicinal plants used in folkloric medicine. Journal of Ethnopharmacology. 2001;74:217–220.
12. Mostafa AA, Al-Askar AA, Almaary KS, Dawoud TM, Sholkamy EN, Bakri MM. Antimicrobial activity of some plant extracts against bacterial strains causing food poisoning diseases. Saudi Journal of Biological Sciences. 2018;25:361–366.
13. Shimada K, Fujikawa K, Yahara K, Nakamura T. Antioxidative properties of xanthin on autoxidation of soybean oil in cyclodextrin emulsion. Journal of Agricultural and Food Chemistry. 1992; 40:945–948.
14. Benzie IF, Strain JJ. The ferric reducing ability of plasma (FRAP) as a measure of antioxidant power: The FRAP assay. Analytical Biochemistry. 1996;239:70–6.

15. Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C. Antioxidant activity applying an improved ABTS radical cation decolorization assay. Free Radical Biology and Medicine. 1999;26(9-10): 1231-1237.
DOI: 10.1016/s0891-5849(98)00315-3
16. Siddhuraju P, Becker K. The antioxidant and free radical scavenging activities of processed cowpea (*Vignaunguiculata* (L.) Walp.) seed extracts. Food Chemistry. 2007;101(1):10-19.
17. Marcocci JJ, Maguire MT, Droy-Lefaix. The Nitric oxide scavenging property Ginkgo biloba extract EGb76. Journal of Biochemical and Biophysical Research Communications. 1994;201: 748-755.
18. Okoli CO, Akah PA, Onuoha NJ, Okoye TC, Nwoye AC, Nworu CS. Acanthus montanus: An experimental evaluation of the antimicrobial, anti-inflammatory and immunological properties of a traditional remedy for furuncles. BMC Complementary and Alternative Medicine. 2008;8:27.
DOI:https://doi.org/10.1186/1472-6882-8- 27
19. Banerjee S, Chanda A, Adhikari A, Das A, Biswas S. Evaluation of phytochemical screening and anti-inflammatory activity of leaves and stem of *Mikania*scandens (L.) Wild. Annals of medical and health sciences research. 2014;4(4):532–536.
DOI:https://doi.org/10.4103/2141-9248.139302
20. Dabhade A, Patel P, Patil U. Proteinaceous protease inhibitor from *Lawsoniainermis*: Purification, characterization and antibacterial activity. Natural Product Communications. 2013;8(10):1467-1470.
21. Anosike CA, Obidoa O, Ezeanyika LU. Membrane stabilization as a mechanism of the anti-inflammatory activity of methanol extract of garden egg (*Solanum aethiopicum*) DARU Journal of Pharmaceutical Sciences. 2012;20(1):76.
22. Ranila LG, Kwon YI, Apostoldis E, Shetty K. Phenolic Compounds: Antioxidant activity and *in vitro* inhibitory potential against key enzymes relevant for hyperglycemia and hypertension of commonly used medicine plants, herbs and spices in Latin America. Bioresource Technology. 2010;101(12): 4676-4689.
23. Kumar S, Pandey AK. Chemistry and biological activities of flavonoids: An overview. The Scientific World Journal; 2013.
Available:https://doi.org/10.1155/2013/162750
24. Chaves JC, Souza MC, Silva Da, Lachos-Perez LC, Torres-Mayanga D, Machado PC, Forster-Carneiro APD, Vázquez-Espinosa T, González-de-Peredo M, AV González-de-Peredo. Barbero GF, Rostagno MA. Front. Chem; 2020.
Available:https://doi.org/10.3389/fchem.2020.507887
25. Zhang X, Shen Y, Prinyawiwatkul W, King JM, Xu Z. Comparison of the activities of hydrophilic anthocyanins and lipophilic tocols in black rice bran against lipid oxidation. Food chemistry. 2013; 141(1): 111–116.
DOI: 10.1016/j.foodchem.2013.03.034
26. Habtemariam S. The chemical and pharmacological basis of yerba maté (Ilex paraguariensis A.St.-Hil.) as potential therapy for type 2 diabetes and metabolic syndrome. Medicinal Foods as Potential Therapies for Type-2 Diabetes and Associated Diseases. 2019;943-983.
DOi- 10.1016/B978-0-08-102922-0.00024-9
27. Ogbole OO, Ayeni FA, Ajaiyeoba EO. In vitro Antibacterial screening of methanol extracts of three *Combretum* species against seven strains of methicillin resistant *Staphylococcus aureus.* Nigerian Journal of Pharmaceutical Research. Combretum. 2016;12(2):149-154.
28. Tungmunnithum D, Thongboonyou A, Pholboon A, Yangsabai A. Flavonoids and Other phenolic compounds from medicinal plants for pharmaceutical and medical aspects: An overview. Medicines (Basel, Switzerland). 2018;5(3):93.
Available:https://doi.org/10.3390/medicines5030093

Peer-review history:
The peer review history for this paper can be accessed here:
https://www.sdiarticle5.com/review-history/102805