The Journal of Phytopharmacology 2019; 8(3): 124-128
Online at: www.phytopharmajournal.com

Research Article

ISSN 2320-480X
JPHYTO 2019; 8(3): 124-128
May- June
Received: 07-05-2019
Accepted: 26-06-2019

DOI: 10.31254/phyto.2019.8307

Afanyibo Y-G
Laboratory of Physiology-Pharmacology, Faculty of Sciences (FDS), Center for Research and Training on Medicinal Plants (CERFOPLAM), University of Lomé (UL), Togo

Esseh K
Laboratory of Physiology-Pharmacology, Faculty of Sciences (FDS), Center for Research and Training on Medicinal Plants (CERFOPLAM), University of Lomé (UL), Togo

Idoh K
Laboratory of Physiology-Pharmacology, Faculty of Sciences (FDS), Center for Research and Training on Medicinal Plants (CERFOPLAM), University of Lomé (UL), Togo

Koudouvo K
Laboratory of Physiology-Pharmacology, Faculty of Sciences (FDS), Center for Research and Training on Medicinal Plants (CERFOPLAM), University of Lomé (UL), Togo

Agbonon A
Laboratory of Physiology-Pharmacology, Faculty of Sciences (FDS), Center for Research and Training on Medicinal Plants (CERFOPLAM), University of Lomé (UL), Togo

Gbeassor M
Laboratory of Physiology-Pharmacology, Faculty of Sciences (FDS), Center for Research and Training on Medicinal Plants (CERFOPLAM), University of Lomé (UL), Togo

Correspondence:
Afanyibo Y-G
Laboratory of Physiology-Pharmacology, Faculty of Sciences (FDS), Center for Research and Training on Medicinal Plants (CERFOPLAM), University of Lomé (UL), Togo
Email: yaogameli@yahoo.fr

Toxicity and antioxidant activity of *Syzygium aromaticum, Mondia whitei, Carissa spinarum* and *Caesalpinia bonduc*

Afanyibo Y-G, Esseh K, Idoh K, Koudouvo K, Agbonon A, Gbeassor M

ABSTRACT

Caesalpinia bonduc, *Mondia Whitei*, *Carissa spinarum* and *Syzygium aromaticum* are four medicinal plants used alone or in combination in the production of traditional liquors in Togo to treat infertility, impotence and infections. They were selected on the basis of an ethnobotanical survey among Togolese traditional medicine practitioners to be evaluated for their toxicological effects and antioxidant properties. The phytochemical extraction was carried out on the powders of the roots, leaves and seeds of *C. bonduc*, the roots and leaves of *M. whitei*, the roots of *C. spinarum* and the leaves of *S. aromaticum*. The toxicity of hydroethanolic and aqueous extracts of each plant organ were checked *in vitro* on shrimp larvae and in antioxidant tests. The toxicity test showed that, the seven hydroethanolic extracts had their LC_{50} between 0.20 and 1.85 mg/ml and were all above 0.1 mg/ml, LC_{50} threshold set for *in vitro* toxicity tests. Concerning antioxidant activity, the DPPH antiradical test showed that the fourteen extracts had it IC_{50} between 38.21 to 96.10 µg/mL, higher than 7.79 µg/mL which is the IC_{50} of quercetin used as a standard drug. The results showed that the extracts had antioxidant activity and were not toxic to shrimp larvae *in vitro*. However, toxicity studies will be further evaluated *in vivo* in animals to prove their safety and their use in traditional medicine in Togo.

Keywords: Medicinal plants, extraction, toxicity, antioxidant.

INTRODUCTION

People's interest in herbal medicine has grown steadily over the years. This is understood by many positive aspects such as its diversity, availability, its low cost, effectiveness and its lesser side effects [1]. However, in low-income countries where most of the population engaged in herbal medicine, the use of plants remains traditional [2]. Scientific knowledge about biological properties of active compounds and the doses to be administered are ignored; and most of the time this situation leads both to treatment failures and toxicological problems.

Indeed, *S. aromaticum (Myrtaceae), M. whitei (Periplocaceae), C. spinarum* (Apocynaceae) and *C. bonduc*, plants of the family of Fabaceae are medicinal plants traditionally used in Togo, alone or in combination, in alcoholic infusions to treat infertility, sexual impotence and infections [3]; but toxicological effects of these plants are unknown by traditional practitioners. Studies have been conducted on these four plants regarding to their biological aspects such as toxicity [4, 5] and antioxidant activity [6, 7, 8]; however, further studies are needed to expand their scientific data to prove their efficacy and safety in view for their inclusion in the traditional pharmacopoeia.

In this context this study was conducted to evaluate the preliminary toxicity and antioxidant property of these medicinal plants used in the preparation of the traditional liquors in Togo.

MATERIAL AND METHODS

Framework of study

The Laboratory of Physiology-Pharmacology of the Faculty of Sciences (FDS) at the University of Lomé (UL) served as the main framework for this study. Phytochemical screening was conducted at Laboratory of Organic Chemistry and Natural Substances (LABCOSNat) FDS/UL. Toxicity tests on shrimp larvae were carried out at the Institute for Research in Traditional Medicine and Pharmacopoeia (IREMPT) in Porto-Novo, Benin.

Plant material

The plant material consists of the roots, leaves and seeds of *Caesalpinia bonduc*, roots and leaves of *Mondia whitei*, fruits of *Syzygium aromaticum* and roots of *Carissa spinarum*.

Fruits of *S. aromaticum* and roots and leaves of *M. whitei* were collected in the village of Tabligbo at the geographic location (6.591041, 1.499805). Roots, leaves and seeds of *C. bonduc* were collected in the village of Wogba (6.283090, 1.509755). Finally, the roots of *C. spinarum* were harvested in the village of Kévé (6.430294, 0.930963). These plants were harvested in October 2017 and forwarded to the Physiology and Pharmacology of FDS/UL. The identification was done in Botany Department where the specimen were deposited in Herbarium under the voucher specific numbers TOGO15347 (*S. aromaticum*), TOGO15350 (*M. whitei*), TOGO15368 (*C. bonduc*) and TOGO15351 (*C. spinarum*).

Shrimp larvae

Brine shrimp larvae, *Artemia salina* LEACH, were used for the preliminary non-clinical toxicity study (larval toxicity *in vitro*). Their incubation was made in seawater collected in the Atlantic Ocean on the coast of Cotonou in Benin.

Extraction

50 g of vegetable powder were macerated with 500 mL of ethanol-water mixture (50:50) for 24 hours with stirring before being filtered. The operation was repeated twice on the extraction residue in 300 mL of the same solvent. For the aqueous extract, a decoction was obtained from 50 g of vegetable powder brought to a boil in 500 mL of distilled water for 30 min. The macerated and decocted were filtered successively on cotton and Whatman No. 40 filter paper. Each filtrate was then evaporated in vacuum at 40 ° C using the Rotavapor R-100 (Buchi, Switzerland). The extracts were stored at -20 ° C until use.

Phytochemical Screening

The major families of secondary metabolites were investigated using classical methods of characterization. Tannins and polyphenols were detected by the $FeCl_3$ test and Stiasny reagent; flavonoids by the cyanidin reaction; the saponins by the foam test; quinones by the Bornträger test and finally alkaloids by the Mayer and Dragendorf tests [9, 10].

Determination of polyphenolic compounds

The total polyphenol content was determined by the Folin-Ciocalteu method then expressed in µg of gallic acid equivalent per mg of extract (µg EAG / mg). Total flavonoids were determined by the $AlCl_3$ method then expressed in µg of quercetin equivalent per mg of extract (µg EQ / mg) [11].

Antioxidant activities: DPPH test

It was carried out using the free radical scavenging method generated by DPPH * (2,2-diphenylpicrylhydrazyl) [11]. A range of 10 concentrations of each extract (0-100 µg / mL) was prepared in the appropriate solvent. A volume of 0.25 mL of the solutions of each extract (solvent alone for the blank) or standard (Quercetin) was added to 1.5 mL of the DPPH solution. The mixture was vigorously homogenized and then incubated at room temperature (25 ± 2 ° C). After 30 minutes incubation in the dark, the absorbance of each concentration of the extracts studied was measured at the wavelength λ = 517 nm using the UV-visible spectrophotometer (Genesys, USA). The percent inhibition of the DPPH radical was calculated by the following equation: % of inhibition = (A0 - AE /A0) x 100 where A0 is the absorbance of the reagent blank, and AE is the absorbance of the tested extracts or standard. The tests were carried out in triplicate.

***In vitro* toxicity test**

The principle of this test is based on the survival of shrimp larvae in seawater in the presence of the test solution. It is carried out according to the method of Michael *et al.* (1956) [12] and Houngbeme *et al.* (2014) [13]. The eggs of *Artemia salina* are incubated in 1000 mL Erlenmeyer flask containing seawater for a period of 48 hours at laboratory temperature until young larvae hatch. A series of extract solutions were then prepared for testing at variable and progressive concentrations. A defined number of larvae (16 larvae) were introduced into the extract and control solutions and subjected to a soft agitation for 24 hours. The count was done under a microscope. In the case of deaths in the control medium, the data are corrected by the Abbott formula: % deaths = [(test-control) / control] × 100.

Data analysis

Graph Pad Prism 6 software was used for data analysis and processing. The results were presented at the threshold $\alpha = 0.05$.

RESULTS AND DISCUSSION

Toxicity

Table 1 summarizes the different values of the half-lethal concentrations (LC_{50}) obtained on the shrimp larvae.

Table 1 : Lethal concentrations 50%

Extract	LC_{50} (mg / mL)
EHRCB	0.49 ± 0.03
EHFCB	0.38 ± 0.01
EHGCB	1.85 ± 0.05
EHRMW	0.42 ± 0.05
EHFMW	0.87 ± 0.02
EHRCS	1.42 ± 0.01
EHFSA	0.20 ± 0.04

The values are expressed in mean ± standard deviation. EHRCB: Hydroethanolic extract of *C. bonduc* roots, EHFCB: Hydroethanolic extract of *C. bonduc* leaves, EHGCB: Hydroethanolic extract of *C. bonduc* seeds, EHRMW: Hydroethanolic extract of the *Mondia* roots *whitei*, EHFMW: Hydroethanolic extract of *Mondia* leaves *whitei*, EHRCS: Hydroethanolic extract of *C. spinarum* roots, EHFSA: Hydroethanolic extract of the fruits of *S. aromaticum*. CL_{50}: Lethal concentration 50%.

The LC_{50} threshold is 0.1 mg / mL: the extract is considered nontoxic for $LC_{50} \geq 0.1$ mg / ml and toxic for $LC_{50} < 0.1$ mg / mL [12, 13]. The analysis in Table 1 shows that the LC_{50} of the extracts tested ranged from 0.2 to 1.85 mg / mL; this suggests that the extracts were not toxic *in vitro* to shrimp larvae at the applied doses because the LC_{50} obtained were above the established limit (0.1 mg / mL).

C. bonduc

The LC_{50} of *C. bonduc* extracts were also greater than 0.1 mg / mL and therefore did not cause any toxicity on shrimp larvae *in vitro*. Acute toxicity studies performed *in vivo* by Mlozi *et al.* (2017) [14] have previously revealed that at $LD_{50} > 2000$ mg / kg body weight, extracts of the plant did not cause any change in mice behavior. This confirms the non-toxic nature of the extracts of *C. bonduc* observed on shrimp larvae *in vitro*.

M. whitei

Oloro *et al.* (2015) [15] showed that the aqueous extract of *M. whitei* roots did not show toxicity at a single dose of 5000 mg / kg; but in 90 days they noted moderate toxicity from the extract. Acute toxicity performed by these authors is consistent with that given by *in vitro* extracts on the shrimp larvae of this study.

C. spinarum

The primary toxicity test showed that hydroethanolic extract of *C. spinarum* was not toxic at the dose of 50 mg /mL on shrimp larvae. LC_{50} roots and leaves were greater than fixed LC_{50} (0.1 mg / mL). However, acute toxicity studies in rats by Ibrahim *et al.* (2011) [16] showed that ethanolic extracts and flavonoids fraction of the plant could be moderately toxic at a dose greater than 646.23 mg / kg. They demonstrated that the ethnoanolic extract was toxic at the dose of 1000 mg / kg of body weight.

S. aromaticum

The preliminary toxicity test showed that *S. aromaticum* extract did not exert toxicity *in vitro* on shrimp larvae. On the other hand, the studies carried out by some authors revealed that the essential oils have a cytotoxic activity [17] and a moderate toxicity (0.25 g / kg / day for 30 days) on the genitals of the rabbits: spermicide in the males and anovulation by alteration of the ovaries in females [18]. These discrepancies in results can be explained by the nature of the extracts used and the doses applied.

The primary toxicity results that were undertaken in this study showed that the four extracts did not exert any toxicity *in vitro* on shrimp larvae. While some results are in agreement with previous studies, others are divergent. The primary toxicity results obtained with *C. bonduc* extracts were confirmed by previous results which showed that even *in vivo*, this plant did not exert toxicity at the dose studied. However, the other three plants showed subacute or acute toxicity *in vivo* with the previous work of the above authors. Considering the results of this study and the previous work, we recommend that these plants, which have long been used in the preparation of medicinal infusions (liquor) by traditional practitioners in Togo, should be consumed with moderation, especially with regard to *M. whitei, C. spinarum* and *S. aromaticum.*

This study is worth pursuing *in vivo* to determine the toxicity and to establish non-toxic doses of each plant extract studied.

Screening phytochemical

Table 2: Phytochemical compounds

Secondary metabolites	RCB	FCB	GCB	RMW	FMW	RSC	FSA
Alkaloids	+	+	+	+	+	+	+
Catholic tannins	-	-	+	-	-	-	+
Gallic tannins	+	+	+	+	+	+	+
Flavonoids	-	+	+	+	+	+	+
Anthocyanins	-	+	+	+	+	+	+
Leucoanthocyanins	-	-	-	-	-	-	-
Quinones	-	-	-	-	-	-	-
Saponosides	+	+	+	+	+	+	+
Cyanogenic derivatives	-	-	-	-	-	-	-
Mucilage	+	+	+	+	+	-	+
Coumarins	+	-	-	-	-	-	+
Reducing compounds	-	+	+	+	+	+	+
Free anthracene derivatives	-	-	-	-	-	-	+
Free glycosides	-	-	-	-	-	-	-
O-Glycosides	-	-	-	-	-	-	-
C-glycosides	-	-	-	-	-	-	-
Cardiotonic glycosides	-	-	-	-	-	-	-

+: Presence; -: Absence; RCB: Roots of *Caesalpinia bonduc*; FCB: Leaves of *Caesalpinia bonduc*; GCB: *Caesalpinia bonduc* seeds; RMW: Roots of *Mondia whitei*; FMW: Leaves of *Mondia whitei;* RSC: Roots of *Carissa spinarum*; FSA: Fruits of *Syzygium aromaticum*

Table 2 presents the results of the qualitative phytochemical analysis of extracts of the four plants. Alkaloids, tannins and saponosides were present in all extracts.

The presences of certain phytochemicals that have been identified in the extracts of these four plants have been proven by some authors. Ogunlana *et al.* (2012) [18] have shown that seeds of *C. bonduc* contain alkaloids, saponins, flavonoids, tannins, and cardiac glycosides. These phytochemicals have also been highlighted in our extracts except for cardiac glycosides which have been found to be negative.

Ethanolic extracts of leaves and roots of *M. whitei* studied by Gbadamosi and Aboaba (2016) [8] revealed the presence of alkaloids, flavonoids, saponins, tannins and polyphenols; which is consistent with the results of this study.

According to Fatima *et al*, (2013) [19], the roots of *C. spinarum* were composed of glucosides, D- digitalose carissone, carindone, carinol, odoroside H, digitoxigenin. The carbohydrates found in the roots of this plant were also present in our extract in addition to other phytochemicals.

Finally, Bhowmik *et al.* (2012) [20], had previously reported in the essential oils of *S. aromaticum* the presence of triterpenes, tannins and flavonoids that were revealed in the leaves of the plant during our phytochemical screening.

Quantitative phytochemical analysis was used to determine the phenols and total flavonoids contained the in extracts of the four plants in gallic acid and quercetin equivalent.

Table 3: Phenolic compounds and antiradical activity of extracts

Extracts	DPPH IC_{50} (µg / mL)	Total polyphenols (mg EAG / g extract)	Total flavonoids (mg EQ / g extract)
EARCB	56.41 ± 12.51	18.24 ± 0.54	Undetermined
EHRCB	47.21 ± 3.02	52.33 ± 0.09	2.75 ± 0.71
EAFCB	65.51 ± 15.46	37.81 ± 1.34	5.28 ± 1.43
EHFCB	83.75 ± 4.65	31.73 ± 1.07	3.86 ± 1.42
EAGCB	69.32 ± 13.34	19.57 ± 0.81	0.33 ± 0.04
EHGCB	95.12 ± 0.33	5.69 ± 0.27	3.06 ± 0.85
EARMW	66.95 ± 12.36	45.32 ± 1.48	6.49 ± 1.43
EHRMW	64.16 ± 2.94	5.59 ± 0.13	0.43 ± 0.02
EAFMW	38.21 ± 4.45	51.50 ± 0.53	13.05 ± 0.71
EHFMW	67.16 ± 6.59	18.52 ± 1.21	4.97 ± 0.99
EARCS	75.65 ± 5.02	200.24 ± 3.09	41.61 ± 1.14
EHRCS	96.10 ± 1.11	35.44 ± 0.13	13.35 ± 0.57
EAFSA	72.64 ± 1.92	213.16 ± 1.21	14.86 ± 1.28
EHFSA	86.69 ± 4.44	353.34 ± 2.96	26.88 ± 2.85
Quercetin	7.79 ± 1.66	-	-

Phenolic compounds were present in the right quantity in the 14 extracts tested at amounts between 5.59 and 353.34 mg EAG / g extract. Flavonoids were detected in all extracts except the aqueous extract of *C. bonduc* roots. The values of flavonoids detected in the 14 extracts range from 0.33 and 41.61 mgEQ / g of extract (Table 3).

Antioxidant activity

The results of radical scavenging tests presented in Table 2 show that all the four plants have antioxidant properties. The inhibitory concentrations 50% (IC_{50}) of the extracts tested were significantly higher than the IC_{50} of quercetin used as a standard. These data prove that extracts of *C. bonduc*, *M. whitei*, *C. spinarum* and *S. aromaticum* have a good anti-radical activity and can therefore be used to trap free radicals generated by oxidative stress or during the body's immune defense against pathogens and xenobiotics. Indeed, it has been shown that oxidative stress, which appears when the endogenous antioxidant system is overwhelmed, contributes to the appearance of numerous pathologies such as cardiovascular and neuro-degenerative diseases [21, 22]. Thus, the intake of antioxidants exogenous would therefore be necessary to help the body to manage free radicals in order to prevent or delay the onset of oxidative stress and related diseases. In addition, other studies have shown that natural antioxidants are more protective than synthetic antioxidants [23]. Plants that contain natural antioxidants will thus be best placed. This antioxidant activity of these four plants observed *in vitro* could be due to the action of certain phytochemicals which are the phenolic compounds. In fact, phytochemical tests proved the presence of phenolic compounds and total flavonoids in the four plants. This could justify the antioxidant activity observed.

Since *C. bonduc*, *M. whitei*, *C. spinarum* and *S. aromaticum* are used to treat infertility and sexual impotence on the one hand and on the other hand *S. aromaticum* especially used against infections, the antioxidant effect of these four plants would be beneficial for consumers to limit the neuro-degeneration that causes these pathologies.

CONCLUSION

This study evaluated the *in vitro* toxicity and antioxidant activity of *C. bonduc*, *M. whitei*, *C. spinarum* and *S. aromaticum*, four medicinal plants that have long been used as an infusion to treat infertility, sexual impotence and infections in traditional medicine in Togo. The results showed that the four plants possess an antiradical activity that can be used in the treatment of those pathologies to reduce the oxidative stress induced. Preliminary toxicity tests conducted on shrimp larvae did not show *in vitro* toxicity. However, these results are not enough to prove the non-toxic nature of the extracts tested especially as some data are at odds with other previous works. This study should be continued to determine *in vivo* in animals, toxicity of each plant in order to decide on their safety in traditional medicine.

Acknowledgments

Our thanks go to LABCOSNat UL and IREMPT in Porto-Novo, Benin.

Conflict of interest

No conflict of interest

REFERENCES

1. Organisation mondiale de la Santé. Stratégie de l'OMS pour la médecine traditionnelle pour 2002-2005, 2002. Genève. 2002.
2. Atchou K. Evaluation du potentiel antifongique de cinq plantes utilisées dans le traitement des mycoses. Bull. Soc. Pathol. Exot., 2013; 106:321. http://www.pathexo.fr/docfiles/07_-_presentation_atchou.pdf
3. Afanyibo YG, Koudouvo K, Esseh K, Agbonon A, Tozo K, Gbeassor M. An ethnobotanical survey of medicinal plants used in the preparation of "Atikédi": local alcoholic beverages commonly consumed in Lomé Togo. European Scientific Journal. 2018; 14:1-17.
4. Hegde K, Joshi AB. Preliminary phytochemical screening and antipyretic activity of *Carissa spinarum* root extract. Der Pharmacia Lettre, 2010; 2:255-260.
5. Noorani AA, Gupta K, Bhadada K, Kale MK. Protective effect of methanolic leaf extract of *Caesalpinia bonduc* (L.) on gentamicin-induced hepatotoxicity and nephrotoxicity in rats. Iranian Journal of Pharmacology and Therapeutics. 2011; 10:21-30.
6. Gupta M, Mazumder UK, Sambath Kumar R, Sivakumar T, Gomathi P, Rajeshwar Y. Antioxidant defense system induced by a methanol extract of *Caesalpinia bonducella* in rat liver. Pharmaceutical Biology, 2005; 43:411-419.
7. Atawodi SE, Atawodi JC, Pfundstein B, Spiegelhalder B, Bartsch H, Owen R. Assessment of the polyphenol components and *in vitro* antioxidant properties of *Syzygium aromaticum* (L.) Merr. & Perry. Electronic Journal of Environmental, Agricultural & Food Chemistry. 2011; 10:1970-1978.
8. Gbadamosi IT, Aboaba SA. Essential oil constituents and *in vitro* antimicrobial activity of the root of *Mondia whitei* (Hook. F.) Skeels (Periplocaceae). Journal of Pharmacognosy and Phytotherapy. 2016; 8:163-167.
9. Harbone JB. Phytochemical methods. Chapman and Hall, ed. New York, 1973.
10. Houghton P, Raman A. Laboratory handbook for the fractionation of natural extracts. Chapman & Hall 1st ed. London, UK, 1998.

11. Bahorun T, Gressier B, Trotin F, Brunet C, Dine T, Luyckx M, *et al*. Oxygen species scavenging activity of phenolic extracts from hawthorn fresh plant organs and pharmaceutical preparations. Arzneimittel-forschung, 1996; 46:1086-1089.
12. Michael AS, Thompson CG, Abramovitz M. *Artemia salina* as a test organism for bioassay. Science (New York, NY), 1956; 123:464.
13. Houngbeme AG, Gandonou C, Yehouenou B, Kpoviessi SD, Sohounhloue D, Moudachirou M, *et al*. Phytochemical analysis, toxicity and antibacterial activity of Benin medicinal plants extracts used in the treatment of sexually transmitted infections associated with hiv/aids. International Journal of Pharmaceutical Sciences and Research. 2014; 5:1739-1745.
14. Mlozi SH, Chacha M, Peter R. Antimycobaterial and antibacterial activities of extracts from *Caesalpinia bonduc* (L.) Roxb. American Journal of Research Communication. 2017; 5:27-36.
15. Oloro J, Tanayen JK, Barbra K, Imanirampa L, Waako P, Bajunirwe F, *et al*. Toxicity of four herbs used in erectile dysfunction; *Mondia whiteii*, *Cola acuminata*, *Urtica massaica*, and *Tarenna graveolensin* male rats. African Journal of Pharmacy and Pharmacology. 2015; 9:756-763.
16. Prashar A, Locke IC, Evans CS. Cytotoxicity of clove (*Syzygium aromaticum*) oil and its major components to human skin cells. Cell Proliferation, 2006; 39:241-248.
17. Alzahid AA, Faris J, Kodiar OS. Histopathological study of the toxic doses of Clove Oil *Syzygium aromaticum* "on Ovaries of female rabbits." IOSR Journal of Agriculture and Veterinary Science. 2015; 8:33-38.
18. Ogunlana OO, Ogunlana OE, Ntube CA, Olagunju JA, Akindahunsi AA. Phytochemical screening and *in vivo* antioxidant activity of ethanolic extract of *Caesalpinia bonduc* (L.) Roxb. Global Research Journal of Pharmaceutical Sciences. 2012; 1:1-5.
19. Fatima A, Singh PP, Agarwal P, Irchhaiya R, Alok S, Verma A. Treatment of various diseases by *Carissa spinarum* L. A promising shrub. International Journal of Pharmaceutical Sciences and Research. 2013; 4:2489-2495.
20. Bhowmik D, Kumar KS, Yadav A, Srivastava S, Paswan S, *et al*. Recent trends in Indian traditional herbs *Syzygium aromaticum* and its health benefits. Journal of Pharmacognosy and Phytochemistry. 2012; 1:13-23.
21. Babbs CF. Oxygen radicals in ulcerative colitis. Free Radical Biology and Medicine, 1992; 13:169-181.
22. Aruoma OI. Free radicals, oxidative stress, and antioxidants in human health and disease. Journal of the American Oil Chemists' Society. 1998; 75:199-212.
23. Zhu YZ, Huang SH, Tan BKH, Sun J, Whiteman M, Zhu YC. Antioxidants in Chinese herbal medicines: a biochemical perspective. Natural product reports. 2004; 21:478-489.

HOW TO CITE THIS ARTICLE

Afanyibo Y-G, Esseh K, Idoh K, Koudouvo K, Agbonon A, Gbeassor M. Toxicity and antioxidant activity of *Syzygium aromaticum*, *Mondia whitei*, *Carissa spinarum* and *Caesalpinia bonduc*. J Phytopharmacol 2019; 8(3):124-128.