



***In vivo* Antidiabetic Activities of Aqueous Extract of *Anchomanes difformis* (Blume) Eng, *Rhizophora racemosa* G. Mey and *Ravenala madagascariensis* Sonn**

**A. K. Camara¹, E. S. Baldé^{1,2*}, M. Haidara³, A. Camara¹, K. Guilavogui⁴,
R. Sanogo^{3,5} and A. M. Baldé^{1,2}**

¹*Département des Sciences Pharmaceutiques et Biologique, CEA-PCMT, University Gamal Abdel Nasser, Conakry, Guinea.*

²*Institute of Research and Development of Alimentary and Medicinal Plants, Dubreka, Guinea.*

³*Faculté de Pharmacie, Université des Sciences des Techniques et des Technologies de Bamako (USTTB), BP 1805, Bamako, Mali.*

⁴*National herbarium of Guinea, Conakry, Guinea.*

⁵*Département de Médecine Traditionnelle de l'Institut National de Recherche en Santé, BP 1746, Bamako, Mali.*

Authors' contributions

This work was carried out in collaboration among all authors. Author AKC designed the study, wrote the protocol, and wrote the first draft of the manuscript. Author ESB wrote the protocol, performed the statistical analysis and review the first draft of the manuscript. Author MH managed the analyses of the study. Author AC managed the literature searches. Author KG collected and identified the plants samples. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/EJMP/2020/v31i1830339

Editor(s):

(1) Dr. Naseem A. Qureshi, National Center of Complementary and Alternative Medicine, Saudi Arabia.

(2) Prof. Marcello Iriti, University of Milan, Italy.

Reviewers:

(1) Diorge J. Marmitt, University of Taquari Valley - Univates, Brazil.

(2) R. Arun Kumar, PRIST Deemed to be University, India.

Complete Peer review History: <http://www.sdiarticle4.com/review-history/62592>

Original Research Article

Received 20 September 2020

Accepted 26 November 2020

Published 14 December 2020

ABSTRACT

Aims: *Anchomanes difformis* (Blume) Eng (Araceae), *Rhizophora racemosa* G. Mey (Rhizophoraceae) and *Ravenala madagascariensis* Sonn (Strelitziaceae) are used in traditional medicine in Guinea for diabetes management. The aim of this work was to test the antidiabetic activity of these plants and to determine their toxicity.

*Corresponding author: E-mail: baldesaid@yahoo.fr;

Material and Methods: Extemporaneous extracts based on these plants were tested for their acute toxicity, their effects in normoglycemic rats and rendered hyper glycemc by the oral route in comparison with the glibenclamide an antidiabetic agent. Swiss albino mice and Male Wistar rats were used respectively for acute oral toxicity and antidiabetic activity.

Results: This study showed that the administration of the 2000 mg / kg dose of dry extracts in mice showed no acute toxicity and adverse effect. At a dose of 400 mg/kg these three plants showed significant hypoglycaemia effect. The average blood glucose levels ranged from 111.2 ± 6.2 to 84.6 ± 6.7 mg/dL ($p < 0.001$) for *A. difformis*, 110.6 ± 6 to 93.2 mg/dL ($p < 0.05$) for *R. racemosa* and 99.6 ± 14.4 to 82.3 mg/dL ($p < 0.05$) for *R. madagascariensis*.

Conclusion: The results obtained on the antidiabetic properties of these three plants corroborates their traditional uses in the management of type 2 diabetes in the Republic of Guinea.

Keywords: Antidiabetic; *Anchomanes difformis*; *Rhizophora racemosa* ; *Ravenala madagascariensi* ; toxicity.

1. INTRODUCTION

Human exploitation of plant diversity in the provision of medicine follows two main strategies: random screening guided by taxonomy where chemotaxonomy draws on the phylogenetic relatedness in the search for identical or similar bioactive compounds, or ethnopharmacological approaches from therapeutic indications of indigenous drugs [1]. Medicinal use of herbal medicines in the treatment and prevention of non-communicable diseases, including diabetes and hypertension, has a long history compared to that of conventional medicine [2]. Due to the side effects of conventional drugs, attention is now focusing on the development of nano-sized herbal drugs and their precise and targeted delivery, which may help to overcome the limitations and complications of conventional drugs used to control diabetes mellitus [3].

Ethnopharmacological surveys have indicated that more than 1,200 plants are used worldwide in traditional medicine for their alleged hypoglycaemic activity [4-12], like *Anchomanes difformis* ENGL, *Anthocleista vogelii* Planch, *Garcinia lancifolia* Roxb, *Ravenala madagascariensis* Sonn, *Zanthoxylum armatum* DC. Management of diabetes using traditional remedies is widespread in Africa in rural as well as urban communities. An increasing number of patients opt for traditional remedies driven by a combination of factors, including financial constraints and/or geographical accessibility of the population to orthodox antidiabetics, the inadequacy of the health care system, ease of accessibility of traditional medicine, indigenous knowledge by community members and the role of traditional healers [13-14].

In the Republic of Guinea, previous ethnobotanical investigations indicated a wide used of plant species in the management of diabetes, particularly type 2 diabetes. And, the relatively low cost of treatment and easier access to medicinal plants could explain the popularity of herbal medicines [15-17].

Among the listed plants *Anchomanes difformis* (Blume) Engl (Araceae), *Rhizophora racemosa* G. Mey (Rhizophoraceae) and *Ravenala madagascariensis* Sonn (Strelitziaceae) feature prominently. *A. difformis*, *R. racemosa* and *R. madagascariensis* are plants with several reported therapeutic effects, and they are used in many parts of Africa for the traditional management of malaria, asthma, diabetes, antimicrobials [18-23].

This study aimed to evaluate the antidiabetic activity of these three plants, on Male Han Wistar rats and their toxic effects on swiss albino mice.

2. MATERIALS AND METHODS

2.1 Herbal Materials

Voucher specimens were collected, and botanical identification was conducted by Karman Guilavogui (a botanist at the National Herbarium of Guinea). The voucher specimens of *Anchomanes difformis* (Blume) Engl. (HNG002577), *Rhizophora racemosa* G. Mey (HNG002578) and *Ravenala madagascariensis* Sonn (HNG002579) were deposited at the National Herbarium of Guinea.

The petioles of *R. madagascariensis* were harvested in the Camayenne area (Conakry); the root bark of *R. racemosa* was harvested in the

Yimbaya area (Conakry), and the tuber of *A. difformis* was harvested in Coyah (50 km from Conakry). The samples were dried in the drying room of the Pharmacognosy Laboratory of the Pharmaceutical and Biological Sciences Training Unit of the University of Conakry after a period of 45 days and were then sprayed.

2.2 Animal Material

Male Wistar rats weighing between 150-215 g were used for the antidiabetic activity evaluation and female Swiss albino mice weighing between 22-28 g were used for the acute toxicity evaluation.

Animals were maintained in standard and constant laboratory conditions (23 – 25 °C and light/dark cycles, i.e., 12/12 h). The animals were fasted for 16 h, weighed and marked, and had free access to water before the start of the tests.

2.3 Methodology

2.3.1 Preparation of plant extracts

Five grams of each powdered sample was infused into 200 mL of boiling distilled water for 15 min. Infusions were filtered and cooled in the refrigerator.

2.3.2 Acute oral toxicity study

The acute oral toxicity study was conducted using test guidelines on acute oral toxicity (test 423) according to the Organization for Economic Cooperation and Development and as described by Alanin [20,22,23,19,18].

Mice were randomly divided into four groups of three female mice and treated by the oral route with a single dose of *A. difformis*, *R. racemosa*, *R. madagascariensis* or water. For *A. difformis*, *R. racemosa*, and *R. madagascariensis* a dose of 2,000 mg/kg (corresponding to 50 mL/kg) was administered, while 20 mL/kg was used for water treatment. Animals were observed for the first 4 h after treatment to record the immediate death and once daily for 14 days to record any manifestation of toxicity.

2.3.3 Evaluation of the antidiabetic activity

2.3.3.1 Assay in normoglycemic rats

Animals were deprived of food for 16 h during the experiment but were allowed free access to

water. They were randomly divided into 4 groups of 5 rats. At time T₀, glycaemia was determined from blood taken from the tails of the rats using an Accu-Chek glucometer. Group 1 received distilled water (10 mL/kg, per os), and groups 2-4 received extemporaneous solutions (10 mL/kg, per os corresponding to 400 mg/kg) of *A. difformis*, *R. racemosa*, and *R. madagascariensis*. Blood samples were taken from the tails of the rats every hour for 3 h (T₁, T₂, and T₃) to determine glycaemia.

2.3.3.2 Oral glucose tolerance tests for evaluation of antihyperglycemic activity

Animals were deprived of food for 16 h during the experiment, but were allowed free access to water. They were randomly divided into 5 groups of 5 rats. At time T₀, glycaemia was determined from blood taken from the tails of the rats using an Accu-Chek glucometer. Glibenclamide (5 mg/kg), distilled water (10 mL/kg) and extemporaneous solutions (10 mL/kg) of *A. difformis*, *R. racemosa*, and *R. madagascariensis* were administered orally 30 min after glucose load (4 g/kg). Blood samples were taken 30 min after the administration of glucose (T 0.5 h) and at 1, 2 and 3 h after the administration of the treatments to determine blood glucose levels.

2.4 Analysis and Expression of Results

Results were expressed in mg / dl as mean ± SD. The statistical analysis was performed by one-way analysis of variance (ANOVA) followed by Student's *t*-test. Significant differences were indicated by *p* values lower than 0.05.

3. RESULTS

3.1 Acute Toxicity

In the acute toxicity study, oral administration of 2000 mg/kg plant extract per group did not induce any changes in behaviour, body weight, or food consumption, and no mortality was observed during the study. The oral LD₅₀ value was estimated superior to 2000 mg/kg plant extract.

3.2 Hypoglycemic activities of aqueous extracts of *A. difformis*, *R. racemosa* and *R. madagascariensis*

In control animals, the average blood glucose level was 98.4±7.4 mg/dL and 85.8±6.7 mg/dL

after the oral administration of 10 mL distilled water, with 0 h values considered the control.

Oral administration of the aqueous extract *A. difformis*, *R. racemosa* and *R. madagascariensis* at the dose of 10 ml/Kg (400 mg/kg) body weight produced significant hypoglycaemia effect (Fig. 1). The average blood glucose levels ranged from 111.2 ± 6.2 to 84.6 ± 6.7 mg/dL (T0 – T3; $p < 0.001$) for *A. difformis*, 110.6 ± 6 to 93.2 mg/dL (T0 – T3; $p < 0.05$) for *R. racemosa* and 99.6 ± 14.4 to 82.3 mg/dL (T0 – T3; $p < 0.05$) for *R. madagascariensis*.

3.3 Antihyperglycemic activities of aqueous extracts of *A. difformis*, *R. racemosa* and *R. madagascariensis*

At the dose of 400 mg per kg body weight (10 ml/Kg), the lowering of blood glucose levels was significant ($p < 0.001$) in experimental animals compared to blood glucose levels in the control

group at the time intervals of 30, 60, 120 and 180 min (Fig. 2).

In all groups, the glucose peak was observed 30 min after hyperglycaemia induction. The mean blood glucose levels obtained were 158.6 ± 18.7 mg/dL 30 min after overload and 81.2 ± 16.8 mg/dL 3 h after overload for the *A. difformis* extract, 138 ± 4.4 mg/dL 30 min after glucose challenge and 105.6 ± 12.3 mg/dL 3 h after administration of the *R. racemosa* extract, and 86.2 ± 8 mg/dL at time zero and 74.2 mg/dL 3 h after glucose overload of the *R. madagascariensis* extract. Excepted for the group1(distilled water), the glycaemia value regression started on time T30 and no significant changes of blood glucose levels were observed in normal-glycemic rats treated with the three plant extracts or glibenclamide at a dose of 5 mg/kg body weight during the 3 h of the experimental period.

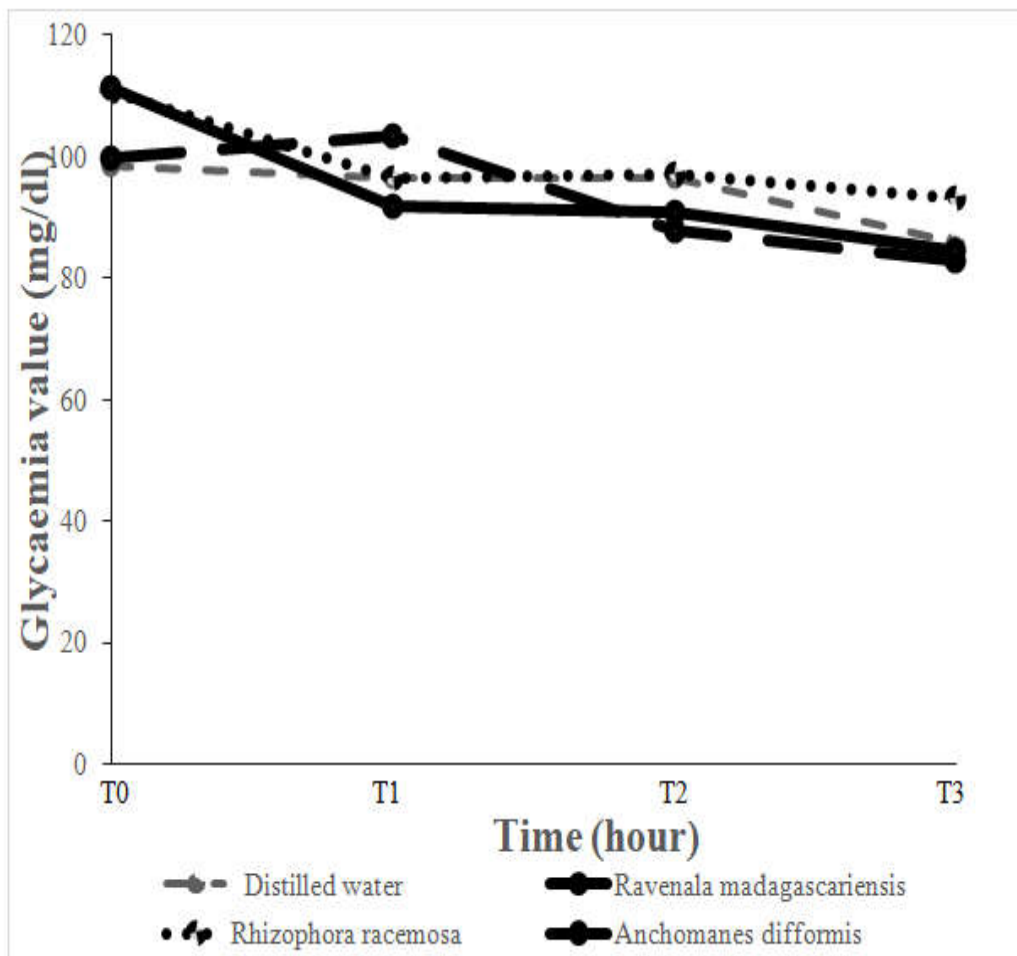


Fig. 1. Hypoglycaemia activity evaluation of aqueous extract *A. difformis*, *R. madagascariensis*, *R. racemosa*

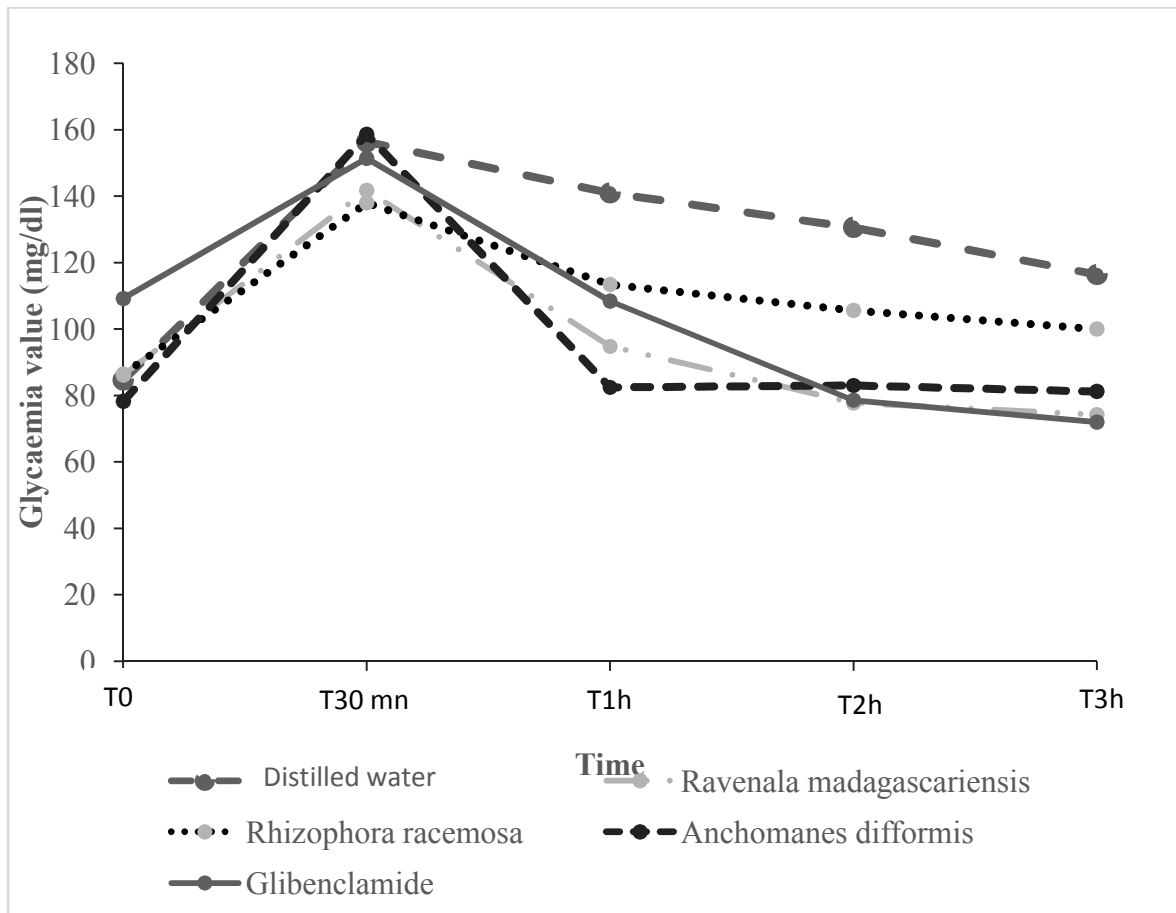


Fig. 2. Antihyperglycaemia activity evaluation of aqueous extract *A. difformis*, *R. madagascariensis*, *R. racemosa* compared to glibenclamide and distilled water

4. DISCUSSION

The observed data suggested that the oral LD50 values of *A. difformis*, *R. racemosa* and *R. madagascariensis* are greater than 2000 mg/kg in Swiss albino mice. Therefore, according to the OECD Globally Harmonized Classification System [24] extracts from *A. difformis*, *R. racemosa* and *R. madagascariensis* are categorized as category 5 (2000 mg/kg < LD50 < 5000 mg/kg) and considered non-toxic orally. The low toxicity of these plants has been confirmed by other authors in various studies [25,26]. We found that the innocuity of *A. difformis*, *R. racemosa* and *R. madagascariensis* is in concordance with the wide use of these plants in traditional medicine [19,21,22,27].

The hypoglycaemic activity of the extemporaneous extract of *A. difformis* revealed an effect on basal glucose at the third hour after administration. We found that glycaemia should be monitored when the plant is used. The aqueous extract of *A. difformis* effectively

decreased blood glucose after hyperglycaemia induction in rats. However, in the control group, hyperglycaemia persisted at the end of the experiment. Our data suggest that *A. difformis* possesses potential antidiabetic activity. In a previous study, ethanolic extracts of the leaves and roots of *A. difformis* at doses of 250 mg/kg and 500 mg/kg demonstrated significant decreases in hyperglycaemia in albino rats [10,11]. Phytochemical screening of *A. difformis* revealed the presence of flavonoids, saponins, and tannins and suggest that the plant is very rich in antioxidant compounds [25]. Antioxidant compounds are responsible of protective roles of vegetables and plant foods against diabetes and other neurodegenerative diseases.

The hypoglycaemia activity of the extemporaneous extract of *R. racemosa* revealed an effect on the basal glucose at the third hour after administration. This shows that glycaemia should be monitored when the plant is used. The data revealed that the *R. racemosa* aqueous extract induced a significant decrease

in hyperglycaemia after induction, thus corroborating the potential antidiabetic activity of the plant. The antidiabetic activity of *R. racemosa* reported in Douala was 250 mL of decoction 3 times daily: 2 g of stem bark per kg of body weight should be boiled in 2 L of water for 15 min to drink [28]. Revathi et al. reported that the flowers and leaves of *R. racemosa* contain compounds with antidiabetic activity [29]. Cinchonain Ia, Cinchonain Ib, Catechin-3-O-rhamnopyranoside, Epicatechin, Iyoside and nudiposide were found to be in *R. racemosa* and could be associated to the hypoglycemic effect of *R. racemosa* [30].

The hypoglycaemia activity of *R. madagascariensis* evaluated at the dose of 10 mL/kg caused a non-significant effect on basal glucose at the third hour after administration. At this same dose, the antihyperglycaemia activity was also evaluated, and we found a significant decrease in blood glucose levels in the rats for the first hour, and the antihyperglycaemic effects persisted until the end of the experiment. Priyadarsini et al. reported that administration of both ethanolic and aqueous leaf extracts of *R. madagascariensis* produced a significant reduction in blood glucose levels in a dose-dependent manner. The ethanolic extract was found to be more effective than the aqueous extract, which may be due to the higher solvent extraction ability of ethanol compared with water [6]. Cycloartanol, a triterpene with antidiabetic activity was found to be in *R. madagascariensis* [31].

5. CONCLUSION

Achieving correct diabetes care without side effects is a challenge for the research community. This study confirmed the therapeutic efficacy of aqueous extracts of the tubercle of *A. difformis* (Blume) the root bark of *R. racemosa*, and the petioles of *R. madagascariensis* in hyperglycaemic rats. The results obtained on the antidiabetic properties of these three plants corroborates their traditional uses in the management of type 2 diabetes in the Republic of Guinea.

CONSENT

It is not applicable.

ETHICAL APPROVAL

Animal experiments were conducted at the Department of Traditional Medicine of the

National Institute of Public Health Research in Bamako, according to the local ethical committee of the institution.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Leonti M, Stafford GI, Cero MD, Cabras S, Castellanos ME, Casu L, et al. Reverse ethnopharmacology and drug discovery. *J Ethnopharmacol.* 2017; 198:417–31.
2. Choudhury H, Pandey M, Hua CK, Mun CS, Jing JK, Kong L, et al. An update on natural compounds in the remedy of diabetes mellitus: A systematic review. *J Tradit Complement Med.* 2018;8(3):361–76.
3. Amjad S, Jafri A, Sharma AK, Serajuddin M. A novel strategy of nanotized herbal drugs and their delivery in the treatment of diabetes: Present status and future prospects. *J Herb Med.* 2019;100279.
4. Anyanwu GO, Iqbal J, Khan SU, Zaib S, Rauf K, Onyeneke CE, et al. Antidiabetic activities of chloroform fraction of *Anthocleista vogelii* Planch root bark in rats with diet- and alloxan-induced obesity-diabetes. *J Ethnopharmacol.* 2019;229: 293–302.
5. Salgueiro ACF, Folmer V, Bassante FEM, Cardoso MHS, da Rosa HS, Puntel GO. Predictive antidiabetic activities of plants used by persons with diabetes mellitus. *Complement Ther Med.* 2018;41: 1–9.
6. Priyadarsini SS, Vadivu R, Jayshree N. In vitro and *In vivo* antidiabetic activity of the leaves of *Ravenala madagascariensis* Sonn, on alloxan induced diabetic rats. *J Pharm Sci Technol.* 2010;2(9):312–7.
7. Rynjah CV, Devi NN, Khongthaw N, Syiem D, Majaw S. Evaluation of the antidiabetic property of aqueous leaves extract of *Zanthoxylum armatum* DC. using *in vivo* and *in vitro* approaches. *J Tradit Complement Med.* 2018;8(1):134–40.
8. Chakraborty R, Roy S, Mandal V. Assessment of traditional knowledge of the antidiabetic plants of Darjeeling and Sikkim Himalayas in the context of recent phytochemical and pharmacological advances. *J Integr Med.* 2016;14(5):336–58.

9. Ghosh A, Banik S, Amin MN, Ahmed J. Evaluation of antinociceptive, antihyperglycemic, and membrane stabilizing activities of *Garcinia lancifolia* Roxb. *J Tradit Complement Med.* 2018; 8(2):303–7.
10. Aderonke SO, Ezinwanne AJ. Evaluation of the anti-diabetic activity of ethanol extract of *anchomanes difformis* (araceae) leaves in albino rats. *Int Res J Pharm.* 2015;6(2):90–3.
11. Adeyemi O, Makinwa TT, Uadia RN. Ethanol extracts of roots of *anchomanes difformis* ENGL roots as an Antihyperglycemic Agent in Diabetic Rats. *Chem J.* 2015;1(3):68–73.
12. Baliga MS, Fernandes S, Thilakchand KR, D'souza P, Rao S. Scientific validation of the antidiabetic effects of *Syzygium jambolanum* DC (Black Plum), a traditional medicinal plant of India. *J Altern Complement Med.* 2013;19(3):191–7.
13. Alamin MA, Yagi AI, Yagi SM. Evaluation of antidiabetic activity of plants used in Western Sudan. *Asian Pac J Trop Biomed.* 2015;5(5):395–402.
14. Kesari AN, Kesari S, Singh SK, Gupta RK, Watal G. Studies on the glycemic and lipidemic effect of *Murraya koenigii* in experimental animals. *J Ethnopharmacol.* 2007;112(2):305–11.
15. Balde ES, Camara AK, Traore MS, Balde NM, Megalizzi V, Pieters L, et al. The hypoglycemic and cytotoxic activity of the leave extract of *Combretum glutinosum* Perr ex DC. *Journal of Pharmacognosy and Phytochemistry.* 2019;8(4):2230–7.
16. Baldé N, Youla A, Baldé M, Kaké A, Diallo M, Baldé M, et al. Herbal medicine and treatment of diabetes in Africa: An example from Guinea. *Diabetes Metab.* 2006;32(2):171–5.
17. Diallo A, Traore MS, Keita SM, Balde MA, Keita A, Camara M, et al. Management of diabetes in Guinean traditional medicine: an ethnobotanical investigation in the coastal lowlands. *J Ethnopharmacol.* 2012;144(2):353–61.
18. Bero J, Hannaert V, Chataigné G, Hérent M-F, Quetin-Leclercq J. In vitro antitrypanosomal and antileishmanial activity of plants used in Benin in traditional medicine and bio-guided fractionation of the most active extract. *J Ethnopharmacol.* 2011;137(2):998–1002.
19. Oghale O-U, Idu M. Phytochemistry, anti-asthmatic and antioxidant activities of *Anchomanes difformis* (Blume) Engl. leaf extract. *Asian Pac J Trop Biomed.* 2016; 6(3):225–31.
20. Oyeyemi IT, Akinseye KM, Adebayo SS, Oyetunji MT, Oyeyemi OT. Ethnobotanical survey of the plants used for the management of malaria in Ondo State, Nigeria. *South Africa J Bot.* 2019; 124:391–401.
21. Wekhe NS, Ogbamgba KO, Oboh CC. Preliminary investigation of the effect of *Rhizophora racemosa* (mangrove) feed additive on broiler performance. *Afr J Biotechnol.* 2007;6(16):1963–5.
22. Suroowan S, Pynee KB, Mahomoodally MF. A comprehensive review of ethnopharmacologically important medicinal plant species from Mauritius. *South Afr J Bot.* 2019;122:189–213.
23. Tchicailat-Landou M, Petit J, Gaiani C, Miabangana ES, Kimbonguila A, Nzikou J-M, et al. Ethnobotanical study of medicinal plants used by traditional healers for the treatment of oxidative stress-related diseases in the Congo Basin. *J Herb Med.* 2018;13:76–90.
24. Martinez-Mayorga K, Marmolejo-Valencia AF, Cortes-Guzman F, García-Ramos JC, Sánchez-Flores EI, Barroso-Flores J, et al. Toxicity Assessment of Structurally Relevant Natural Products from Mexican Plants with Antinociceptive Activity. *J Mex Chem Soc.* 2017;61(3):186–96.
25. Aliyu AB, Ibrahim MA, Musa AM, Musa AO, Kiplimo JJ, Oyewale AO. Free radical scavenging and total antioxidant capacity of root extracts of *anchomanes difformis* engl. (araceae). *Acta Pol Pharm.* 2013; 7(1):115–21.
26. Ukoima H, Ikata M, Pepple GA. Control of *Lasiodiplodia theobromae* (PAT) on *Rhizophora racemosa* using plants extracts. *Am J Biotechnol Mol Sci.* 2013; 3(1):1–7.
27. Ataman JE, Idu M. Renal effects of *Anchomanes difformis* crude extract in wistar rats. *Avicenna J Phytomedicine.* 2015;5(1):17–25.
28. Tsabang N, Nanga N, Fokunang Tembe E, Agbor GA. Herbal Medicine and Treatment of Diabetes in Africa: Case Study in Cameroon. *Diabetes Case Rep.* 2017;01(02).
29. Revathi P, Jeyaseelan ST, Thirumalaikolundusubramanian P, Prabhu

- N. An overview of antidiabetic profile of mangrove plants. *International J Pharm Pharm Sci.* 2014;6(3):1–5.
30. Andrade-Cetto A, Escandón-Rivera SM, Torres-Valle GM, Quijano L. Phytochemical composition and chronic hypoglycemic effect of *Rhizophora mangle* cortex on STZ-NA-induced diabetic rats. *Rev Bras Farmacogn.* 2017; 27(6):744–50.
31. Ramiarantsoa H, Yao-Kouassi PA, Kanko C, Assi KM, Djakoure AL, Tonzibo FZ. Chemical constituents of the antidiabetic plant *Ravenala madagascariensis*. *Int J Pharm Sci Res.* 2014;5(12):5503–10.

© 2020 Camara et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:

The peer review history for this paper can be accessed here:
<http://www.sdiarticle4.com/review-history/62592>