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Department of Biochemistry
Usmanu Danfodiyo University, Sokoto

Book of Abstracts

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NSBMB Sokoto 2021

THEME

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Venue: University Auditorium, Main Campus, Usmanu Danfodiyo University, Sokoto

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NIGERIAN SOCIETY OF BIOCHEMISTRY
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38th SCIENTIFIC CONFERENCE AND
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SOKOTO 2020
Usmanu Danfodiyo University, Sokoto
Department of Biochemistry

15 May, 2021

Dear Author(s): Umar, M. B., Ogbadoyi, E. O., Kabiru, A. Y., and Mann, A.

ACCEPTANCE LETTER

We are pleased to inform you that after review by the Technical Sub-committee, your abstract titled **"*In vitro* Antidiabetic Activity of Crude Ethanol Extracts of Selected Medicinal Plants"** has been accepted for Poster presentation **(Poster size: horizontal 115 cm, vertical 90 cm)** at the forthcoming 38th Annual Conference of the Nigerian Society of Biochemistry and Molecular Biology (NSBMB) to be hosted by Usmanu Danfodiyo University, Sokoto, Nigeria between the 6th and 10th Jun, 2021.

Accept our warmest regards.

Sincerely,

Prof. R. A. Umar

Chairman, Technical Sub-committee

ED079

. Antidiabetic Effect of Extracts of *Ziziphus mucronata on* Alloxan-Induced Diabetic Rats

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ABSTRACT

Ziziphus mucronata is an important multi-purpose plant species that has been used in African traditional medicine for ages in the treatment of various devastating human infections including diabetes. The current study was designed to investigate the *in vitro* antidiabetic activity and *in-vivo* evaluation of antidiabetic effect in diabetic animal model. Sequential extraction of *Ziziphus mucronata* using n-hexane, acetone, methanol and water were respectively used to access the *in-vitro* antidiabetic effect on α-amylase and haemoglobin glycosylation inhibitions. Standard laboratory methods were used to screen for phytochemicals while acute oral toxicity (LD_{50}) was estimated using up and down procedure. *In-vivo* antidiabetic activity was investigated using alloxan-induced animal model. The results reveal that methanol, acetone and aqueous extracts exhibited dose-dependent increase in percentage inhibition. Phytochemicals detected in *Zizphus mucronata* leaf acetone extract include alkaloids, anthraquinone, glycosides, flavonoids, phenols, saponins, tannins and terpinoids. $LD_{50}$of acetone leaf extract was estimated to be greater than 5000mg/kg as no mortality was recorded after 14 days. Sub-chronic (21 days) administration of *Zizphus mucronata* acetone leaf extract exhibited significant reduction ($p<0.05$) in fasting blood glucose at 400 mg/kg body weight compared with normal control.In conclusion, the present findings suggest that the most potent extract (acetone) exhibited blood glucose lowering activity. This validates the traditional claim of *Z. mucronata* in the treatment of diabetes.

Keywords: Phytochemicals, *Ziziphus mucronatam Dabetes, Glucose*

ED080

In vitro Antidiabetic Activity of Crude Ethanol Extracts of Selected Medicinal Plants

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ABSTRACT

The incidence of diabetes mellitus has increased globally in the last two decades and there is no cure for it yet. However, several plants have been reported to be used locally for its management. This study was aimed at evaluating the *in vitro* anti-diabetic activity of the crude ethanol extracts of fourteen (14) medicinal plants used locally for diabetes therapy. This was carried out using non-enzymatic methods (*in vitro* haemoglobin glycosylation inhibitory assay and *in vitro* assay of glucose uptake by yeast cells). The highest percentage inhibition of haemoglobin glycosylation (96.68±0.34 %) was observed for crude ethanol extract of *Calotropis procera* at concentration 20 mg/mL, with no significant difference ($p > 0.05$) when compared to *Anogeissus leiocarpus* (95.02±0.22 %), *Khaya senegalensis* (92.55±1.60 %), *Senna occidentalis* (92.12±2.48 %), *D. microcarpum* (91.11±0.38 %) and the standard drug, Acarbose (90.15±0.30 %). The crude ethanol extracts of *Balanites aegyptiaca* (*B. aegyptiaca*), *Khaya senegalensis*, *Mangifera indica*, *Senna podocarpa* and the standard drug (metronidazole) had percentage glucose uptake by yeast cells of up to 70 % at the lowest concentration used (0.625 mg/mL) for all the concentrations of glucose tested. However, the highest percentage glucose uptake was observed for ethanol extract of *B. aegyptiaca* (91.76±0.88 %) at concentration 20 mg/mL, for 20 mM glucose concentration. It could be concluded that ethanol extracts of all the plants used in this study contain active compounds responsible for inhibiting

haemoglobin glycosylation, as well as enhancing the uptake of glucose in yeast cells. As such, some of them with remarkable activity could be standardised and used as herbal remedies for the treatment of diabetes mellitus.

Keywords: Diabetes mellitus, Inhibition, Haemoglobin, Glycosylation, Yeast cells

ED081

Gastroprotective Activity of Ethanolic Extract of *Raphia mambillensis* Seed on Indomethacin-induced Ulcerated Rats

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ABSTRACT

Raphia mambillensis seed is applied traditionally as a treatment to different ailments including gastrointestinal related disorders by the rural dwellers in Nigeria. The aim of this study is to evaluate the antiulcer activity of ethanol extract of *Raphia mambillensis* seed in ulcerated rats. Animals were pre-treated orally with different doses of ethanolic seed extract of *Raphia mambillensis* and reference drug (ranitidine).The effect of ethanolic extract of *Raphia mambillensis* (EERM) on ulcer index, stomach acidity and volume of gastric contents was evaluated in ethanol-induced ulcerated rat models using indomethacin (10mg/kg body weight).Standard phytochemical methods were used to screen the presence of its phytoconstituents. The result showed that EERM significantly ($P < 0.05$) decreased the severity of gastric mucosal damage caused by indomethacin indicative by a reduction in ulcer index, gastric acidity and gastric volume of treated groups when compared with untreated group and. may be attributed to its phytochemical constituents such tannin, saponins, alkaloid, cardiac glycoside and anthraquinone. *Raphia mambillensis* leaves possess anti-ulcer effect and could be a potential source of anti-ulcer agent for future research.

Keywords: *Raphia mambillensis*, Ranitidine, Mucosal damage, Ulcer index, Indomethacin.

ED082

Phytochemicals and *In vitro* Antioxidant Analysis of Ethylacetate Leaf Extract of *Corchoris olitorius*

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ABSTRACT

Phytochemicals are biologically active chemical compounds founds in plants, which provide health benefits for humans as medicinal ingredients and nutrients. Antioxidants have been found to play a significant role in the body defense against reactive oxygen species. *Corchurus olitorius* leaves are used in traditional medicine for cystitis, dysuria, fever and gonorrhea. The research work investigates phytochemicals and *in vitro* antioxidant activity of ethylacetate extract of *Corchorus olitorus* leaves. The phytochemical analysis, DPPH radical scavenging assay, Reducing power assay, Hydroxyl radical scavenging activity, Hydrogen peroxide assay, total antioxidant activity were carried out by standard methods. The results of the phytochemical screening indicated the presence of flavoniods, terpeniods, tannins, saponins and cardiac glycosides. The antioxidant activities of *C. olitorius* extract (1000 ug/mL) recorded were total antioxidant activity (DPPH) 82.88±0.005%, reducing power assay 2.070+0.0030 Absorbance, Hydroxyl radical scavenging assay 94.66±0.046%, hydrogen peroxide scavenging assay 82.44 ± 0.37% Antioxidant Capacity 5.505+ 1.188 ug/mL. The research revealed that ... possess good *in vitro* radical ...

In vitro Antidiabetic Activity of Crude Ethanol Extracts of Selected Medicinal Plants

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Background

Diabetes mellitus is a metabolic disease characterized by hyperglycaemia, resulting from defects in insulin secretion, insulin action, or both (Wesam *et al.*, 2016). According to the International Diabetes Federation (IDF), an estimated 19 million adults aged 20-79 years were living with diabetes in African Region in 2019 and this is projected to increase by 143 % by 2045, if nothing is done (IDF, 2020). Nigeria recorded over 2.7 million affected adult (aged 20-79 years) and 63,958 deaths resulting from diabetes in 2019 (IDF, 2020). Some of the therapeutic approaches for treating type 2 diabetes are to decrease postprandial hyperglycaemia through inhibition of glycated haemoglobin and enhancing the uptake of glucose for cellular activities. The use of medicinal plants has recently gained popularity all over the world for their efficacy in type 2 diabetes The effects of antidiabetic plants may delay the development of diabetic complications and even assist in correcting the metabolic abnormalities (Chauhan *et al.*, 2010). Plant derived drugs and herbal formulations are frequently considered to be less toxic and free from side effects than synthetic ones (Bahmani-Ahmadi *et al.*, 2016).

Objective

- To evaluate the *in vitro* antidiabetic activity of crude ethanol extracts of selected medicinal plants

Methods

Preparation of haemoglobin

Blood sample was collected from a healthy human volunteer into a blood bottle containing an anticoagulant. Hemolysate was prepared based on the principle of hypotonic lysis (Adisa *et al.*, 2004). The red blood collected was washed thrice with 0.14 M NaCl solution and one volume of red blood cells suspension was lysed with two volumes of 0.01 M phosphate buffer, pH 7.4 and 0.5 volume of CCl_4. The hemolysate was then freed from the debris by centrifugation at 2300 rpm for 15 min at room temperature. The haemoglobin rich fraction i.e. the upper layer was separated and dispensed into sample bottle for storage and refrigerated until required for use.

Inhibition of haemoglobin glycosylation assay

Antidiabetic activity of the extracts was investigated by estimating the degree of non-enzymatic haemoglobin glycosylation, measured colorimetrically at 520 nm. Glucose (2 %), haemoglobin (0.06 %) and gentamycin (0.02 %) solutions were prepared in phosphate buffer of 0.01 M, pH 7.4. 1 mL each of above solutions were mixed. 0.2 to 1 mg/mL of the plant extracts was then added to the mixture.

The mixture was incubated in the dark at room temperature for 72 hrs after which the degree of glycosylation of haemoglobin was measured colorimetrically at 520 nm. Acarbose was used as a standard drug for the assay. Percentage inhibition was calculated as described by Das and Siddhartha (2011).

In vitro assay of glucose uptake by yeast cells

Yeast cells were prepared according to the method of Cirillo (1962). Commercial baker's yeast was washed by repeated centrifugation (3,000 × g; 5 min) in distilled water until the supernatant fluid was clear and a 10 % (v/v) suspension was prepared in distilled water. Various concentrations of extracts (0.625, 1.25, 2.5 and 5 mg/mL) were added to 1 mL of glucose solutions (5, 10 and 25 mM) and incubated together for 10 minutes, at 37 °C. Reaction was started by adding 100 µL of yeast suspension, vortexed and further incubated at 37 °C for 60 min. After 60 min, the tubes were centrifuged (2,500 × g; 5 min) and glucose was estimated in the supernatant. Metronidazole was taken as standard drug. The percentage increase in glucose uptake by yeast cells was calculated using the following formula;

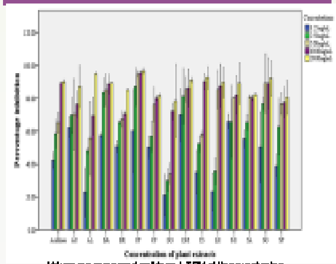
Increase in glucose uptake (%) = [(Abs sample-Abs control)/Abs sample] x 100

Where, Abs control is the absorbance of the control reaction (containing all reagents except the test sample), and Abs sample is the absorbance of the test sample. All the experiments were carried out in triplicates.

Statistical analysis

Data were calculated as mean ± SEM and were analysed statistically using One-way ANOVA followed by Duncan multiple comparison test and values of $p<0.05$ were considered significant. Statistical Package for Social Sciences (SPSS), 20th version was used.

Results



Values are expressed as Mean ± SEM of three replicates

Figure 1: Haemoglobin Glycosylation Inhibitory Activity of Leaf Ethanol Extracts of the Selected Plants at Different Concentrations

Keys: Acarbose = Standard drug, AC = A. conyzoides, AL = A. leiocarpus, BA = B. aegyptiaca, BR = B. rotundata, CP = C. procera, CF = C. fistula, DO = D. oliver, DM = D. mespiliforme, KS = K. senegalensis, LH = L. hastata, MI = M. indica, SA = S. alata, SO = S. occidentalis, SP = S. podocarpa

Table 1: Effect of Ethanol Extracts of the Selected Plant Extracts on Glucose (5 mM) Uptake by Yeast Cells

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Table 2: Effect of Ethanol Extracts of the Selected Plant Extracts on Glucose (10 mM) Uptake by Yeast Cells

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Table 3: Effect of Ethanol Extracts of the Selected Plant Extracts on Glucose (25 mM) Uptake by Yeast Cells

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Discussion

High glucose levels in the body leads to nonenzymatic binding to haemoglobin (haemoglobin glycosylation) which may result in the production of reactive oxygen species which usually plays a key role in many degenerative diseases including diabetes. It is evident from this study that crude ethanol extracts of the leaves of the selected plants demonstrated significant inhibition of haemoglobin glycosylation in a concentration dependent manner and some of the plant extracts demonstrated better inhibitory activity than the standard drug (Acarbose). This further indicates the potential of these plants as agents for antidiabetic drug development and aligns with the reports of Chaudhari *et al.* (2013); Bhukar and Bhise (2013); Nair *et al.* (2013); Sampath *et al.* (2014); Singh and Kumar (2015) and Bhukar *et al.* (2017) who have reported the effectiveness of medicinal plants in decreasing the formation of glucose-haemoglobin complex and thus increasing the amount of free haemoglobin. This study revealed that all the plant extracts increased glucose uptake in yeast cells significantly at various glucose concentrations. The significant phytochemical contents of medicinal plants are suspected to be responsible for the marked increase in percentage glucose uptake with increasing concentration of the extracts and of glucose (Pitchaipillai and Ponniah, 2016). The observed increase in glucose uptake by yeast cells enhanced by the extracts is in agreement with previous reports; *B. aegyptiaca* for example, have been reported by Motaal *et al.* (2012) as well as Mirya *et al.* (2018) to increase *in vivo* muscle basal glucose uptake. Ngo *et al.* (2019) also reported the *in vitro* effectiveness of *M. indica* leaf extracts in L0-2 liver cells

Conclusion

The present study established that all the plant extracts tested exhibited significant haemoglobin glycosylation inhibitory activity as well as enhancing the uptake of glucose by yeast cells. This study directs further researches to evaluate the therapeutic potentialities of the selected plant extracts in the management of postprandial hyperglycaemia and Type 2 diabetes *in vivo*.

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