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Phytochemical Screening and *in vivo* Inhibitory Study of *Telfaira occidentalis* Hook f. Seeds Extract on Alpha Amylase and Alpha Glucosidase of Rats

Godwin Ndarake Enin[1]*, Jude Efiom Okokon[2], Blessing Okon Odokwo[1], Bassey Sunday Antia[1]

[1]Department of Chemistry, Faculty of Science, University of Uyo, Uyo, Nigeria
[2]Department of Pharmacology and Toxicology, Faculty of Pharmacy, University of Uyo, Uyo, Nigeria
*Corresponding author: Godwin Ndarake Enin
E-mail: enin.godwin@gmail.com
Tel. no. +2347025042102

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Abstract

Diabetes mellitus (DM) is a serious metabolic disorder affecting about 425 million people worldwide with estimated 2.2 million deaths annually. The management of diabetes mellitus by the use of orthodox medicines has been unsatisfactory with huge financial burden to the patients in addition to the associated side effects. Therefore, the search for affordable, relatively safe and sustainable materials as alternative to synthetic drugs is inevitable. In this study, seeds of Telfaira occidentalis, a vegetable plant widely consumed in most part of Nigeria was evaluated for its in vivo inhibitory role on alpha amylase and alpha glucosidase enzymes in rats. The seed extracts (150, 300 and 450 mg/kg) were investigated using starch, sucrose, and maltose as substrates and acarbose as reference drug. The seed extract caused significant ($p<0.05$) reduction in blood glucose levels of the treated rats with the various substrates used. Preliminary phytochemical screening of the seed extract showed that alkaloids, flavonoids, saponins, cardiac glycosides, anthraquinones and tannins were present. The findings show that the seed extract of Telfaira occidentalis has inhibitory effects on alpha amylase and glucosidase in rats which, may be attributed to the presence of the phytochemical compounds.

1.0. Introduction

Diabetes mellitus (DM) is a serious metabolic disorder affecting about 425 million people worldwide with estimated 2.2 million deaths annually [1]. Findings show that, over 151 million cases of diabetes were documented in 2000, and about 415 million in 2015 [2]; [3]. This figure was projected to reached 700 million by 2045 [4]. In 2014, about 25 million cases of diabetes were reported in Africa [5], 4.7 million were in Nigeria, accounting for 5.7% of the incidence in Africa [3]. The management of diabetes mellitus by the use of orthodox medicines has been unsatisfactory with huge financial burden to the patients in addition to the associated side effects. The use of hypoglycemic sulphonyl urea and antihyperglycemic biguanides such as metformin and phenformin increase peripheral uptake of glucose, and reduce hepatic glucose output [6]. However, these synthetic compounds pose serious financial burden and side effects over long term administration. Therefore, the search for affordable, relatively safe and sustainable materials as alternative to synthetic counterpart is inevitable. Plant has been an alternative source of potent antidiabetic drugs because it is cheaper, eco-friendly and sustainable- a large number have been screened for possible antidiabetic activity [6]. Inhibition of alpha amylase and alpha glucosidase enzymes, which are enzymes involved in glucose absorption, has been one of the antidiabetic modes of action of drugs, of which, plants are used in the management of the disorder.

Telfaira occidentalis Hook is a fluted pumpkin of the Cucurbitaceae family widely consumed as food in Nigeria [7]. It is a popular vegetable all over Nigeria, especially in the Niger-Delta region and the Eastern part of the country; the leaves, stem and seeds are used to prepare different kinds of meals [8]. The seeds are very nutritious and are eaten roasted or boiled. Leaves are good sources of proteins, vitamins (B-complex), minerals, fatty acids (linoleic and oleic acids), and fibers [9]. Biological activities reported in the plant include: antioxidant, antidiabetic, immunodulatory, anticancer, antiinflammatory, antiplasmodial, antibacterial, hepatoprotective effects and male fertility activity [10]. The median lethal dose of the seed extract was estimated to be 3.46g/kg [11]. Phytochemical studies of the root extract have shown the presence of alkaloid, flavonoid, tannins, terpenes, saponin, and cardiac glycosides [12]. Despite the reported antidiabetic action of *Telfaira occidentalis*, its *in vivo* effect on alpha amylase and glucosidase has not been elucidated. In this study, we investigated the phytochemical constituents of the ethanol seed extract and its inhibitory activity on alpha-amylase and alpha-glucosidase of Wister rats.

2.0. Materials and Methods

2.1. Materials

The materials used include *Telfaira occidentalis* powdered seed extract, oral gastric gavage, weighing balance, gloves, scissors, glucometer and strips (fine test), distilled water, acarbose (Aldrich sigma, USA; standard drug), stirrer, beakers, 1ml syringe, starch, sucrose, maltose (Aldrich sigma, USA)**.**

2.1.1. Plant collection

Fresh seeds of *Telfaira occidentalis* (Figure 1) were purchased from a market (Udua Okpopo) in Etinan L. G. A, Akwa Ibom State, Nigeria, in June, 2023. The seeds were identified in the Department of Botany and Ecological Study, University of Uyo, Uyo, Nigeria.

2.1.2. Extraction

Fresh seeds of the plant were washed, dried on laboratory table for 2 weeks and reduced to powder. Powdered sample (700 g) was macerated in 95% ethanol (4400 mL) for 72 hours. The liquid filtrate obtained was concentrated *in vacuo* at 40˚C to completely remove the ethanol. The yield was calculated, and the extract was stored in a refrigerator at 4˚C until used for experiment.

2.1.3. Phytochemical Screening

Phytochemical screening for the presence of saponins, tannins, flavonoids, alkaloids, cardiac glycosides and anthraquinones was conducted following previously reported standard procedures [13].

2.1.4. Animals

Albino Wistar rats (120 -135 g) of either sex maintained at animal house of the Department of Pharmacology and Toxicology, Faculty of Pharmacy, University of Uyo, Nigeria were used for the study. The animals were housed in standard cages and were maintained on a standard pelleted feed (Guinea feed) and water *ad libitum*. Permission and approval for animal studies were obtained from the College of Health Sciences Animal Ethics committee, University of Uyo.

2.1.5. Alpha-Amylase inhibitory study

Thirty Wistar rats were divided into 6 groups of 5 rats each. The rats in all groups were fasted for 18 h and fasting blood glucose concentration was first taken at 0 min before administration. Group I, as the normal control, received distilled water (10 mL/kg). Group II rats were orally administered starch at 2 g/kg body weight (orally with distilled water as vehicle) and distilled water (10 mL/kg) simultaneously. Rats in group III were administered starch (2 g/kg) and the standard drug (acarbose) at 100 mg/kg simultaneously. Groups IV, V and VI were administered simultaneously, starch (2 g/kg) and *Telfaira occidentalis seed* extract at 150, 300 and 450 mg/kg

respectively. All administrations were done orally and blood glucose concentration was monitored at 30, 60, 120 and 180 min [14]. The blood glucose level was used to assess the effect of extract on the enzyme activity.

2.1.6. In vivo alpha glucosidase inhibition study

The procedure as described above was used for this study but with sucrose and maltose used as substrates [14].

2.1.7. Blood Glucose Determination

Drops of blood from tip of rats' tails were dropped on stripes and glucose concentration was measured using a glucometer according to manufacturer's specifications (fine test). The glucometer works with the following principle; the blood sample is exposed to a membrane covering the reagent pad (strip), which is coated with an enzyme (glucose oxidase, glucose dehydrogenase). The reaction causes a colour change and the intensity of this change is directly proportional to the amount of glucose in the blood sample. Light from an LED strikes the pad surface and is reflected to a photodiode, which measures the light intensity and converts it to electrical signals. An electrode sensor measures the current produced when the enzyme converts glucose to gluconic acid. The resulting current is directly proportional to the amount of glucose in the sample [15].

2.1.8. Statistical Analysis

Data obtained were analyzed statistically using one –way ANOVA followed by Tukey-Kramer multiple comparison post test using InstatR Graphpad software, (San Diego, USA). Differences between means were considered significant at $p< 0.05$ and very significant at $p < 0.001$.

Figure 1: *Telfaira occidentalis* Hook dry seeds

3.0. Results

3.1. Extraction

The extraction yield was 75.95 g. This is calculated to afford percentage yield of 10.85% w/w.

3.2. Phytochemical screening

The results of the phytochemical screening revealed the presence of saponins, tannins, flavonoids, alkaloids, cardiac glycosides and anthraquinones (Table 1).

Table 1: Preliminary phytochemical screening of *T. occidentalis seed extract*

Parameter	Result
Alkaloid	+
Saponins	+
Tannin	+
Anthraquinone	+
Flavonoid	+
Reducing sugar	ND
Cardiac glycosides	+
Steroids	ND

Key: + present; - absent; ND not determine

3.3. In vivo alpha amylase and glucosidase inhibition assay

Administration of starch (2 g/kg) to fasted rats caused varying percentages of increase in blood glucose levels of the treated animals after 30 mins. The percentages were starch (39.42%), *T. occidentalis* seed extract-treated groups (31.35-37.05%), and acarbose-treated group (17.97%). These increases were reduced after 60 min BGL of 24.31, 26.39 and 14.59 were respectively recorded for 150, 300 and 450 mg/kg of the extract treated groups. All the extract-treated groups had their BGL reduced to a normal level at 180 min. Also, co-administration of the starch with acarbose prominently inhibited the rise in the blood glucose concentrations (Table 2).

Administration of maltose (2 g/kg) to fasted rats caused varying percentages of increase in blood glucose levels of the treated animals after 30 mins. There was a 53.60% increase in blood glucose level 30 min following maltose administration in the control group. However, 50.60 - 57.10 % increases were observed in the *T. occidentalis* seed extract-treated groups. At 60 min, groups treated with 150, 300 and 450 mg/kg extract had their BGL reduced to normal and sustained throughout the duration of the study (Table 3).

Administration of sucrose (2 g/kg) to fasted rats caused varying percentages of increase in blood glucose levels of the treated animals after 30 mins. The percentages were sucrose (73.18%), *T. occidentalis* seed extract-treated groups (38.79 - 58.22%), and acarbose-treated group (3.37%). These increases were reduced to normal after 60 min with only the group treated with 300 mg/kg of the seed extract having BGL increment of 3.51 %. All the groups treated with the seed extract had their BGL reduced to a normal level at 120 min and this was sustained throughout the study. Also, co-administration of the starch with acarbose prominently inhibited the rise in the blood glucose concentrations (Table 4).

Table 2: Effect of ethanol seed extract of *Telfaira occidentalis* on Blood Glucose Level of rat after oral administration of starch load

TREATMENT	DOSE	BLOOD GLUCOSE LEVEL mg/dL IN MIN				
	mg/kg	0 min	30 min	60 min	120 min	180 min
Control (Normal saline)	-	100.00±4.25	88.33±1.85	92.33±4.25	89.0±4.35	87.33±3.84
Starch	2000	87.50±4.97	122.0±9.95[b](39.42)	107.50±3.70[a](22.85)	103.0±5.67(17.71)	90.50±9.63(3.42)
Acarbose	100	72.33±2.69	85.33±12.97(17.97)	80.33±7.21(11.06)	74.0±1.00(2.30)	72.33±8.68(0)
Extract	150	91.50±3.62	121.0±9.44[c](32.24)	113.75±4.78(24.31)	100.0±5.33(9.28)	88.25±10.21
	300	98.50±10.57	135.0±4.65[b](37.05)	124.50±10.13(26.39)	114.25±10.40(15.98)	92.95±1.89()
	450	92.50±6.29	121.50±4.73(31.35)	106.0±3.39(14.59)	95.0±4.06(2.70)	91.25±5.07()

Data is expressed as MEAN ± SEM. Significant at [a]$p<0.05$, [b]$p< 0.01$, when compared to control (n=6). Values in parenthesis are percentage increases in blood glucose concentrations compared to 0 min in the same group.

Table 3: Effect of ethanol seed extract of *Telfaira occidentalis* on Blood Glucose Level of rat after oral administration of maltose load

TREATMENT	DOSE	BLOOD GLUCOSE LEVEL mg/dL IN MIN				
	mg/kg	0 min	30 min	60 min	120 min	180 min
Control (Normal saline)	-	86.00±11.53	87.66±7.12(1.93)	87.66±7.62(1.93)	91.0±7.50(5.81)	80.00±6.02
Maltose		86.75±2.52	133.25±7.21[a](53.60)	125.75±1.65(44.95)	99.50±2.90(12.75)	88.0±1.68(1.44)
Acarbose	100	85.34±1.36	88.22±1.10(3.37)	86.0±2.20(0.77)	84.26±1.14[a]()	82.28±2.26()
Extract	150	86.25±2.85	135.50±4.83(57.10)	84.25±8.67()	67.75±7.81()	63.0±5.43()
	300	87.0±2.73	131.25±11.81(50.86)	83.50±3.88()	75.25±1.60()	71.25±2.52()
	450	85.50±3.50	130.0±6.39(52.04)	73.0±1.22()	74.75±2.49()	66.75±3.14()

Data is expressed as MEAN ± SEM, Significant at [a]$p<0.05$, [b]$p< 0.01$, when compared to control (n=6). Values in parenthesis are percentage increases in blood glucose concentrations compared to 0 min in the same group.

Table 4: Effect of ethanol seed extract of *Telfaira occidentalis* on Blood Glucose Level of rat after oral administration of sucrose load

TREATMENT	DOSE	BLOOD GLUCOSE LEVEL mg/dL IN MIN				
	mg/kg	0 min	30 min	60 min	120 min	180 min
Control (normal saline)	-	100.00±4.25	88.33±1.85	92.33±4.25(1.80)	89.0±4.35(1.55)	87.33±3.84(3.98)
Sucrose	2000	79.25±2.05	137.25±4.07^{b}(73.18)	125.50±2.72^{b}(58.35)	95.25±3.32^{b}(20.18)	86.0±4.91^{a}(8.51)
Acarbose	100	85.34±1.36	88.22±1.10(3.37)	86.0±2.20(0.77)	84.26±1.14^{a}()	82.28±2.26()
Extract	150	87.0±4.37	120.75±6.50^{a}(38.79)	85.75±1.43^{a}()	74.0±3.08^{a}()	77.50±4.19^{a}()
	300	85.25±3.56	120.25±7.59^{b}(41.05)	88.25±1.03^{a}(3.51)	75.50±2.66^{a}()	74.0±5.55()
	450	81.75±2.52	129.35±4.92^{b}(58.22)	89.50±2.21^{b}(7.75)	74.25±4.95^{b}()	73.50±2.66^{a}()

Data is expressed as MEAN ± SEM, Significant at $^{a}p<0.05$, $^{b}p< 0.01$, when compared to control. (n=6). Values in parenthesis are percentage increases in blood glucose concentrations compared to 0 min in the same group.

3.4. Discussion

This work investigated the phytochemical constituents present in the seed extract of *Telfaira occidentalis* Hook and its effects on alpha amylase and alpha glucosidase in rats. Preliminary phytochemical investigation revealed: Alkaloids, flavonoids, cardiac glycosides, saponins, tannins and anthraquinones in the seed extract of the plant. The seed extract was found to inhibit increases in blood glucose concentration following starch administration independent of the dose. It has been reported that complete digestion of dietary polysaccharides like starch is achieved by the combined action of α-amylases and α-glucosidase enzymes [16]. The α-amylase enzyme digests α-bonds of the α-linked polysaccharides yielding disaccharides, like maltose, which are further reduced to monosaccharides by membrane bound α-glucosidase enzymes [16; 17]. Inhibitions of these enzymes delay the digestion of ingested carbohydrates thereby resulting in a small rise in blood glucose concentrations following carbohydrate meals as was observed in this study. As a target for managing Type 2 diabetes mellitus, many medicinal plants have been reported to possess α-amylase and α- glucosidase inhibitory potential [18; 19]. Similarly, the seed extract significantly inhibited blood glucose rises when co-administered with maltose, glucose and sucrose. Acarbose, the standard drug used in this study significantly inhibited blood glucose rise when co-administered with starch, maltose and sucrose. However, the inhibitory activity of acarbose on the enzymes was stronger than that of the seed extract. The results of this study support the antidiabetic activity earlier reported on the seed extract [20] and further suggest the involvement of inhibitory effects on alpha glucosidase and amylase as one of the modes of antidiabetic activity of the leaf extract. The inhibitory activities of plant extracts are linked to their phytochemical constituents. The root extract of *T. occidentalis* has been reported to be rich in flavonoids, terpenes, tannins amongst others [12]. Results of our phytochemical screening of the seed extract also revealed the presence of saponins, tannins, alkaloids, flavonoids and cardiac glycosides. These compounds have been previously reported to inhibit alpha glucosidase and alpha amylase activities [21; 22]. Phenols have been reported to inhibit alpha amylase and alpha glucosidase [23]. Also, polyphenolic compounds from plants are known to cause several effects on the biological systems which include enzymes inhibitions [24; 25]. The phenolic compounds are known to be strong metal ion chelators and protein precipitation agents forming insoluble complexes with proteins as well as acting as biological oxidants [26]. The presence of the polyphenolic compounds in the seed ethanolic extract of this plant may suggest that their inhibitory potential on α-amylase and the membrane-bound intestinal α-glucosidase enzymes. The presence of these phytochemicals in the extract may have contributed to the observed activity in this study and therefore explains the antidiabetic mechanism of the seed of *T. occidentalis*.

4.0. Conclusion

The results of this study suggest that inhibition of alpha amylase and alpha glucosidase enzymes maybe one of the modes of antidiabetic activity of the seed extract of *Telfaira occidentalis* which may be attributed to the activities of its phytochemical constituents.

Competing Interest

The authors declare that they have no competing interests.

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References

[1] World Health Organisation (2018). Premature mortality. https://gateway.euro.who.int/en/indicators/h2020_1-premature-mortality/ (Retrieved on 27th February 2022).
[2] International Diabetes Federation (2015). *IDF Diabetes Atlas.* 7th Edition. International Diabetes Federation, Brussels, 145p.
[3] Adeloye, D., Ige, J. O., Aderemi A. V., Adeleye, N., Amoo, E. O., Auta, A. and Oni, G. (2017). Estimating the prevalence, hospitalisation and mortality from type 2 diabetes mellitus in Nigeria: a systematic review and meta-analysis, *British Medical Journal*, 7: e015424.
[4] Saeedi, P., Petersohn, I., Salpea, P., Malanda, B., Karuranga, S., Unwin, N., Colagiuri, S., Guariguata, L., Motala, A.A., Ogurtsova, K., Shaw, J., Bright, D. and Williams, R. (2019). Global and regional diabetes prevalence estimates for 2019 and projections for 2030 and 2045: Results from the International Diabetes Federation. Diabetes Atlas, 9th edition. *Diabetes Research and Clinical Practice* 157: 107843.
[5] World Health Organisation (2016). *Global Report on Diabetes*. World Health Organization, Geneva, 145p.
[6] Deshmukh, C. D., Jain, A., & Nahata, B. Diabetes mellitus: a review. *Int. J. Pure Appl. Biosci*, 2015; *3*(3), 224-230. www.ijpab.com
[7] Agbede, J. O., Adegbenro, M., Onibi, G. E., Oboh, C., & Aletor, V. A. (2008). Nutritive evaluation of Telfairia occidentalis leaf protein concentrate in infant foods. *African Journal of Biotechnology*, 7(15): 2721-2727. http://www.academicjournals.org/AJB.
[8] Okpiabhele, U. U. A., & Adegbegi, A. J. (2023). Telfairia Occidentalis Hook f. Mitigates Carbon-Tetrachloride-Induced Nephrotoxicity in Wistar Rats. *Journal of Science and Technology Research*, *5*(2). https://doi.org/10.5281/zenodo.8010075,
[9] Oboh, G., & Akindahunsi, A. A. (2004). Change in the ascorbic acid, total phenol and antioxidant activity of sun-dried commonly consumed green leafy vegetables in Nigeria. *Nutrition and Health*, *18*(1), 29-36.
[10] Eseyin, O. A., Sattar, M. A., & Rathore, H. A. (2014). A review of the pharmacological and biological activities of the aerial parts of *Telfairia occidentalis* Hook. f. (Cucurbitaceae). *Tropical Journal of Pharmaceutical Research*, *13*(10), 1761-1769. http://dx.doi.org/10.4314/tjpr.v13i10.28
[11] Ebong, A. S., Eseyin, O. A., Etim, E. I., Okokon, J. E., Anah, V. U., Attih, E. E., & Charles, G. E. (2020). *Telfairia occidentalis* Potentiates the Antiplasmodial Activity of Artemisinins and Amodiaquine Combination Therapy. *Anti-Infective Agents*, *18*(2), 152-159. https://doi.org/10.2174/2211352517666190206160812
[12] Enin, G. N., Antia, B. S., Shaibu, S. E., & Nyakno, I. (2023). Comparison of the chemical composition, nutritional values, total phenolics and flavonoids content of the ripe and unripe *Solanum nigrum* Linn. Fruits from Nigeria. *World Journal of Pharmacy and Pharmaceutical Sciences. 12(8)1-18.* DOI: 10.20959/wjpps20238-25410.
[13] Gidado, A., Watafua, M., Sa'ad, R. S., Tagi, H. S. and Abdullahi, S. (2019). Alpha-amylase, maltase and sucrase inhibitions by aqueous leaf extracts of *Anacardium occidentale* (Anacardiacea) and *Piliostigma reticulatum* (Caesalpiniaceae) in rats. *Tropical Journal of Natural Product Research.* 3(6): 210-215. Https://doi.org/10.26538/tjnpr/v3i6.5.
[14] Okokon JE, Ekpo A., Eseyin O. A. (2009). Evaluation of i*n vivo* antimalarial activities of ethanolic leaf and seed extracts of *Telfairia occidentalis. Journal of Medicinal Food*.12 (3): 649 - 653
[15] World Health Organization (2011). Glucose analyser. Core medical equipment information. www.who.int/medical_devices/en/index.html. Accessed 5 June, 2019.
[16] Kalra, S. (2014). Alpha Glucosidase Inhibitors. *The Journal of Pakistan Medical Association*. 64(4): 474-476.
[17] Alongi, M. and Anese, M. (2018). Effect of coffee roasting on in vitro α-glucosidase activity: Inhibition and mechanism of action. *Food Research International*. 111: 480-487. Https://doi.org/10.1016/j.foodres.2018.05.061
[18] Ibrahim, M. A., Koorbanally, N. A. and Islam, M. S. (2014). Anti-oxidative activity and inhibition of key enzymes linked to type 2 diabetes (α-glucosidase and α-amylase) by Khaya senegalensis. *Acta Pharmaceutica*. 64(3): 311-324. Https://doi.org/10.2478/acph-2014-0025
[19] Esimone, C. O., Okonta, J. M. and Ezugwu, C. O. (2001). Blood sugar lowering effect of *Anacardium occidentale* leaf extract in experimental rabbit model. *Journal of Natural Remedies.* 1(1): 60–63.
[20] Eseyin, O. A., Ebong, P., Eyong, E. U., Umoh, E., & AwofIsayo, O. (2010). Comparative hypoglycaemic effects of ethanolic and aqueous extracts of the leaf and seed of *Telfairia occidentalis*. *Turk J Pharm Sci*, *7*, 29-34.
[21] Proença, C., Freitas, M., Ribeiro, D., Oliveira, E. F., Sousa, J. L., Tome, S. M., Ramos M. J., Silva, A., Fernandes P. A. and Fernandes, E. (2017). α-Glucosidase inhibition by flavonoids: an in vitro and in silico structure–activity relationship study. *Journal of Enzyme Inhibition and Medicinal Chemistry*. 32(1): 1216-1228. Https://doi.org/10.1080/14756366.2017.1368503.

[22] Su, J. and Tang, Z. (2019). Effects of (-)-epigallocatechin gallate and quercetin on the activity and structure of α-amylase. *Tropical Journal of Pharmaceutical Research.* 18(3): 585-590. Http://dx.doi.org/10.4314/tjpr.v18i3.20

[23] Oboh, G., Olasehinde, T. A. and Ademosun, A. O. (2017). Inhibition of enzymes linked to type-2 diabetes and hypertension by essential oils from peels of orange and lemon. *International Journal of Food Properties.* 20(sup1): S586-S594. Https://doi.org/10.1080/10942912.2017.1303709.

[24] Kalita, D., Holm, D. G., LaBarbera, D. V., Petrash, J. M. and Jayanty, S. S. (2018). Inhibition of α-glucosidase, α-amylase, and aldose reductase by potato polyphenolic compounds. *PloS One.* 13(1): e0191025. Https://doi.org/10.1371/journal.pone.0191025.

[25] Funke, I. and Melzig, M. F. (2005). Effect of different phenolic compounds on α-amylase activity: Screening by microplate-reader based kinetic assay. *Die Pharmazie-An International Journal of Pharmaceutical Sciences*. 60(10): 796–797.

[26] Ishnava, K. B. and Metisariya, D M. (2018). In vitro study on α-amylase inhibitory activity of selected ethnobotanical plant extra its and its herbal formulations. *International Journal of Pharmacognosy and Chinese Medicine.* 2(3): 01-10.