T.Sundar Rajan et al /J. Pharm. Sci. & Res. Vol. 9(9), 2017, 1586-1588

ISSN:0975-1459
Journal of Pharmaceutical Sciences and Research
www.jpsr.pharmainfo.in

Phytochemical Evaluation and *In vitro* Antidiabetic Activity of Ethanolic Extract of *Amaranthus tristis Linn.*

T.Sundar Rajan[1], Vijey Aanandhi M[2]*

[1]*Research Scholar, Department of Pharmaceutical Chemistry, School of Pharmaceutical Sciences, Vels University (VISTAS), Chennai, Tamilnadu, India*

[2*]*Department of Pharmaceutical Chemistry and Analysis, School of Pharmaceutical Sciences, Vels University (VISTAS), Chennai,Tamilnadu, India*

Abstract

The digestive enzymes like alpha-glucosidase and alpha-amylase played a vital role in the carbohydrate metabolism. Alpha-glucosidase inhibitors are the saccharides that act as hardies in the small intestine.Antidiabetic therapeuticapproach to reduce the postprandial glucose level in blood by the inhibition of alpha-glycosidase and alpha-amylase enzymes. These can be a major strategy for the management of blood glucose level in the body. The aim of the present study was to investigate the phytochemical biomarker compounds of the ethanolic extract of *Amaranthus tristis Linn* leaves and its in-vitro antidiabetic activity. The result suggests that the presence of bioactive compounds could be responsible for the versatile medicinal properties of this plant including diabetes and the extract exhibit the dose-dependent action by increasing the inhibitory effect on the alpha-glucosidase enzyme and alpha-amylase enzyme. The ethanolic extract of *Amaranthus tristis Linn* leaves study exhibited potent hypoglycemic activity by in vitro studies.

Keywords: Alpha-glucosidase, Alpha-amylase, invitro antidiabetic activity, Hypoglycemic activity *Amaranthus tristis Linn.*

INTRODUCTION:

Diabetes mellitus is a worldwide increasing problem entailing enormous financial burden and medical care policy issues. According to International Diabetes Federation (IDF), the number of individuals with diabetes and its complication in 2019 crossed 366 million, with an estimated 4.6 million deaths every year[1]. The Indian subcontinent has emerged as the capital of this diabetes population. Indians show a significantly higher prevalence of diabetes when compared with several other populations. Asian Indians display a higher insulin level which is an indicator of peripheral insulin resistance[2]. The insulin resistance in Indians is thought to be due to their higher body fat percentage. Excess body fat, lack of physical activity and racial predisposition may explain the prevalence of hyperinsulinemia and increased development of type 2 diabetes in Indians[3].

Diabetes characterized by metabolic deregulation primarily of carbohydrate metabolism, manifested by higher blood glucose level resulting from the defect in insulin secretion, insulin action, or both. Uncontrolled diabetes leads to many complications which are leading to peripheral vascular disease, nephropathy, neuropathy, and retinopathy[4].

According to the World Health Organization (WHO), up to 80% of the population in developing countries uses plants and its products as a traditional medicine for primary health care needs. The WHO has listed 21,000 plants, which used for medicinal purposes around the world. Among these, 2500 species are in India. There are about 800 plants which have been reported to show antidiabetic potential. Vast collections of plant-derived phytoactive principles representing numerous natural bioactive compounds have established their role for possible use in the treatment of diabetes[5].

Amaranthus tristis Linn also called “Slender joy weed" is a genus of about 80 herbaceous evergreens, herbaceous perennial plants growing from 15 - 30cm tall and spreading to form a mat of growth 40 cm wide or more. Folk work practice claim that *Amaranthus tristis Linn* has been used as food material as well as for combating various illness. Plants have been reported to possess phytoactive compounds like Amarantin, Isoamarantin, Betaine, Amino acids, Flavonoids, and Phytosterols to contribute the therapeutic action. It has great medicinal value, as it used as an astringent in diarrhea and dysentery[6].

MATERIALS AND METHODS:

I Collection and Authentication

The fresh plant was collected from Gingee Fort VillupuramDist Tamil Nadu and identified from local farmers and authenticated by Pro.Jayaraman plant anatomy research center Tambaram. The herbarium submitted to the Department of Pharmacognosy, Vel's College of Pharmacy, for further reference. Authentification number PARC/2016/3267.

II. Extraction

The leaves part of plants collected, and cleanly remove impurities, and shade dried, and made course powder with the help of dry mechanical grinder, and passed through sieve number 60. The powdered stem and leaves were extracted using soxhleation method. The power defatted with petroleum ether (40-60°C) and followed by Benzene, Chloroform, and Ethanol. Extracts were evaporated to dryness and perform preliminary phytochemical screenings were carried out by using standards methods.

III In vitro methods employed in antidiabetic studies Inhibition of Alpha-Amylase enzyme

A starch solution (0.1% w/v) obtained from photos, by stirring 0.1g of potato starch in 100 ml of 16 mM of sodium

acetate buffer. Buffer solution Prepare by Pipette out exactly 36.2ml of sodium acetate solution into 100ml of a standard flask and add 14.8ml of glacial acetic acid, make the volume 100ml using distilled water using to form sodium acetate buffer. The enzyme solution prepared by mixing 27.5 mg of alpha-amylase enzyme in 100 ml of distilled water. The colorimetric reagent prepared by mixing with sodium potassium tartrate solution and 3,5 dinitro salicylic acid solution 96 mM. Control, chloroform and ethanol extracts of *Amaranthus tristis Linn* were added to the starch solution and left to react with the alpha-amylase solution under alkaline conditions at 25ºC. The reaction was measured over 3 minutes. The generation of maltose was quantified by the reduction of 3,5dinitro salicylic acid to 3- amino-5- nitro salicylic acid. This reaction is detectable at 540 nm [7].

IV Inhibition of Alpha-Glycosidase enzyme

Inhibitory activity of Alpha-Glycosidase determined by incubating a solution of starch substrate (2 % w/v maltose or sucrose) and 1 ml with 0.2 M Tris buffer pH 8.0 and various concentration of plant extract for 5 min at 37°C. The reaction initiated by adding 1 ml of alpha-glucosidase enzyme (1U/ml) to it followed by incubation for 40 min at 35°C. Then the reaction terminated by the addition of 2 ml of 6N HCl. Then the intensity of the color was measured at 540nm[8&9].

Calculation of 50% Inhibitory Concentration (IC50) The concentration of the plant extracts required to scavenge 50% of the radicals (IC50) calculated by using the percentage scavenging activities at five different levels of the extract. Percentage inhibition (I %) was calculated by I % = (Ac-As)/Ac X 100, . where Ac is the absorbance of the control and As is the absorbance of the sample[10].

RESULTS

The preliminary phytochemical screening tests for the methanol extract of *Amaranthus tristis Linn* leaves revealed the presence of carbohydrates, alkaloids, flavonoids, tannins, steroidal glycosides, and phenols. Ethanol extract has more biomarkers presents compare with benzene and chloroform extract. So ethanolic extract of *Amaranthus tristis Linn* selection of in vitro antidiabetic potential. There was a dose-dependent increase in percentage inhibitory activity against alpha- amylase enzyme. At a concentration of 1.0 ml of ethanol extract showed a percentage inhibition 21% and for 1.0 ml plant extract showed inhibition of 97%

Table 1 shows the results of various Phytoconstituents present in the different extracts of *Amaranthus tristis* Linn. Phytochemical screening of petroleum ether, benzene, chloroform and ethanol extracts exposed The presence of alkaloids, carbohydrate, tannins, terpenoids, flavonoids, phenols, etc., by using various methods. Phytochemical screening showed that the maximum phytoconstituents were present in ethanol.

Table II and Table III showed the present study we evaluated in vitro alpha amylase, and alpha glucosidase activity of crude ethanol extract of *Amaranthus tristis* Linnleaves. The plant showed significant inhibition activity, so further the compound isolation, purification, and characterization which is responsible for inhibiting activity, has to be done for the usage of antidiabetic agent.

Diabetes mellitus is a metabolic disorder with increasing incidence throughout the world. Insulin is a key player in the control of glucose homeostasis. Lack of insulin affects carbohydrate, fat and protein metabolism. Management of diabetes without side effects is still challenging to the medical community.

Table I Phytochemical Analysis 0f Ethanol Extract Of*Amaranthus tristis Linn*.

Phytoconstituents	Different Solvent Extracts			
	Petroleum ether	Benzene	Chloroform	Ethanol
Carbohydrates	-	-	+	+
Glycosides	-	-	+	+
Alkaloids	-	-	-	+
Phytosteroids	+	+	+	+
Flavonoids	+	-	+	+
Protein and amino acids	-	-	-	-
Tannins	-	-	-	-
Saponins	-	-	+	+
Gum and mucilages	-	-	-	+

+ indicates presence - indicates absence.

Table II: In Vitro Antidiabetic Activity of *Amaranthus tristis Linn* Alpha-Amylase Method

The concentration of a sample	% inhibition of chloroform extract	% inhibition of ethanol extract
0.2	16.2	21
0.4	32.4	59
0.6	49.7	65
0.8	61.2	81
1.0	72.3	97
IC_{50}	0.56	0.49

Table III In vitro antidiabetic activity of *Amaranthus tristis Linn* Alpha glucosidase method

concentration of sample	% inhibition of chloroform extract	% inhibition of ethanol extract
0.2	26.2	33.6
0.4	34.4	55.1
0.6	51.1	70.1
0.8	63.2	82.5
1.0	74.6	91.2
IC_{50}	0.56	0.49

It proposed that inhibition of the activity of such alpha-amylase and alpha-glucosidase would delay the degradation of carbohydrate, which would, in turn, cause a decrease in the absorption of glucose..As a result the reduction of postprandial blood glucose level elevated. In the present study, research has been carried out to evaluate the preliminary phytochemical investigation and the potential of methanol extract of *Amaranthus tristis Linn*in inhibiting alpha-glucosidase and alpha-amylase.

CONCLUSION

In this present study, we evaluated in vitro alpha amylase, and alpha glucosidase activity of crude methanol extract of *Amaranthus tristis Linn* leaves. The plant showed significant inhibition activity, so further the compound isolation, purification, and characterization which is responsible for inhibiting activity has to be done for the usage of antidiabetic agent.

ACKNOWLEDGEMENT:

The authors are thankful to Vels University (VISTAS) and its management for providing research facilities and encouragement.

REFERENCES

1. L. K. Keter and P. C. Mutiso. Ethnobotanical studies of medicinal plants used by Traditional Health Practitioners in the management of diabetes in Lower Eastern Province, Kenya. J Ethnopharmacology., 2012: 139, (1) 74–80,
2. H. Dong, N. Wang, L. Zhao, and F. Lu. Berberine in the treatment of type 2 diabetes mellitus: a systemic review and meta-analysis. J Evid-Based Complementary and Alternative Medicine., 2012: 2012,1- 12.
3. World Health Organization, Traditional medicine-growing needs and potential.WHO Policy Perspective on Medicines. 2002:2, 1–12.
4. M. K. Ali, K. M. V. Narayan, and N. Tandon. Diabetes & Coronary heart disease: current perspectives. Indi J Medi Research., 2010: 132,11,584–597.
5. M. A. Banerji, N. Faridi, R. Atluri,. Body composition, visceral fat, leptin, and insulin resistance in Asian Indian men. J Clin Endocrinol Meta., 1999: 84, 137–144.
6. Sundarrajan.T vijeyaanandhi.M.Antidiabetic activity of ethanolic extract of *Alternathera ficodia* Linn in streptozotocin-induced diabetic rats. Int j pharm bio sci., volume 2016:7,177 – 180.
7. R. Manikandan*, A.Vijaya Anand. Phytochemical and in vitro anti-diabetic activity of methanolic extract of *Psidium guajava* leaves. Int.J.Curr.Microbiol.App.Sci ,2013: 2,15-19.
8. Narkhede M. B , P. V. Ajimire1 , A. E. Wagh. In vitro antidiabetic activity of *Caesalpina digyna* (R.) methanol root extract. Asi J Plant Sci and Research, 2011:12, 101-106.
9. Y. V. Sashikanth, P. Aravindkumar, and C. Swarupa. Two-way relation of diabetes mellitus and periodontitisa review. Ann and Esse Dentistry, 2012: 4, 1-10 .
10. M. Modak, P. Dixit, J. Londhe, S. Ghaskadbi, and T. P. A. Devasagayam.Indian herbs and herbal drugs used for the treatment of diabetes. J Clin Biochemi and Nutri 2007: 40 3, 163–173.