Palestinian Medical and Pharmaceutical Journal (PMPJ). 2024; 9(1): 107-124

Investigation of Antidiabetic Activity and pharmacokinetics in Herbo-mineral Ayurvedic Formulation 'Arogyavardhini Gutika in streptozotocin induced wistar rat

*Ruchi Bhattacharya[1], Bina Gidwani[2], Shiv Shankar Shukla[2] & Ravindra Kumar Pandey[3,]**

[1]Research Scholar, Department of Pharmacology, Columbia Institute of Pharmacy, Raipur (C.G.), India. [2]Professor, Department of Pharmaceutical Analysis and Quality Assurance, Columbia Institute of Pharmacy, Raipur (C.G.), India. [3]Professor, Department of Pharmacognosy, Columbia Institute of Pharmacy, Raipur (C.G.), India.

*Corresponding author: ravindraiop@gmail.com

Received: (17/1/2023), Accepted: (18/6/2023), Published: (1/3/2024)

ABSTRACT

Aim: Herbs, spices, and minerals are important therapeutic attributes of ayurvedic formulations. The formulation Arogyavardhini Gutika is mentioned in the essential drug list of the Ayurvedic Formulary of India. It has a claimed safety profile and is used for the treatment of jaundice, leprosy, fever, oedema, obesity, skin disorders, and other hepatic issues. Methods: An *in- vitro* study was performed by alpha-amylase inhibition using starch, iodine, and dinitrosalicyclic acid. An *in-vivo* study was performed using a streptozotocin-induced diabetic rat model. Graded doses of Arogyavardhini gutika (200 and 500 mg/kg) were administered orally for 14 days to normal and streptozotocin-induced type I diabetic rats (45 mg/kg, intravenously). Fasting plasma glucose levels were assessed at different time intervals along with lipid profiles (total cholesterol, triglycerides, low density lipoprotein, and high density lipoprotein levels). The pharmacokinetic parameters (Cmax and AUC) of the ayurvedic formulation were also estimated. Result: Arogyavardhini gutika exhibited dose-dependent inhibition of alpha-amylase. The IC50 value for Arogyavardhini gutika was 101.72 g/ml compared with standard acarbose of 79.50 g/ml as estimated by the starch-iodine method. In the DNSA method, Arogyavardhini gutika exhibited an IC50 value of 131.51 g/ml compared with standard acarbose of 82.86 g/ml. In the present research, *in- vivo* studies indicate that the rise in plasma glucose levels in streptozotocin-induced diabetic rats was lowered by arogyavardhini gutika at both doses (250 mg/kg and 500 mg/kg). However, the effective lowering dose was 500 mg/kg. The Cmax and AUC of the ayurvedic formulation as compared to standard acarbose were low, but the Tmax was found to be similar, i.e., 2 hours. Conclusion: The Ayurvedic formulation Arogyavardhini Gutika showed potential antidiabetic activity in a dose-dependent manner. Thus, it may be used as an alternative therapy for the treatment of diabetes mellitus.

Keywords: Arogyavardhini Gutika, Diabetes Mellitus, Acarbose, Starch Iodine Method, Lipid Profile, AUC, Dinitrosalicyclic Acid Method.

INTRODUCTION

Diabetic mellitus is a chronic metabolic disorder characterized by high blood glucose levels (hyperglycemia). High urinary glucose excretion, well-known as glycosuria, is also a prime symptom in DM. It is identified as one of the deadliest diseases in the world that can only be controlled but not cured. Diabetes mellitus results from the malfunction or insufficient production of insulin, which lowers glucose intake and decreases glycogen synthesis. By raising the level of glucose in the blood, this leads to hyperglycemia [1]. The WHO expert committee has listed diabetes as one of its recommendations that traditional methods of treatment for diabetes should be more thoroughly investigated. Different polyherbal anti-diabetic Ayurvedic formulations available in the market are dihar, diabet, diasol, Diabecon, and Karmin Plus [2, 3]. The fact that there is very little scientific support for ayurvedic formulations is what prevents more people from using them. According to the Ayush (Ayurvedic, Yoga, Naturopathy, Unani, Siddha, and Homoeopathy) department of the Government of India, it is urgent that scientific proof be presented. Ayurvedic formulations are prepared with natural ingre-

dients and do not contain any chemical or synthetic components in their entire processing or manufacturing. Padmaja et al., in the year 2022, explored and investigated the standardization parameters and conducted an analysis of the physicochemical parameters of Arogyavardhini gutika/vati [22].

The formulation Arogyavardhini gutika /vati is herbo-mineral formulation mentioned in the essential drug list of ayurvedic formulary of India. Since ancient times, this traditional Ayurvedic remedy has been used to cure jaundice, skin conditions, and other liver ailments. It is considered to be effective and safe. Leprosy, fever, oedema, obesity, jaundice, and other hepatic problems are treated with it [4, 21]. The ingredients of Arogyavardhini gutika are listed in Table 1[20]. Some components of Arogyavardhini Vati, namely *Picrorrhiza kurroa [*5], *Terminalia chebula [6]*, *Terminalia bellerica [7]*, *Emblica officinalis [8]*, and *Guggulu [9]*, are recognized to exhibit hypolipidemic properties. This formulation has already been prescribed for antidiabetic activity for a long time by ayurvedic practitioners in different regions of India, yet there is no documentary evidence for its effect on reducing glucose levels. Therefore, it has been selected to evaluate its antidiabetic action.

Table (1): Ingredient of arogyavardhini gutika.

S. No	***Ingredient***	***Quantity***
1	Shuddha Parada (Herbal purifiedMercury)	1 part
2	Shuddha Gandhaka (Herbal purified Sulphur)	1 part
3	Loha Bhasma (Ash prepared from Iron)	1 part
4	Abhraka Bhasma (Purified and processed Mica)	1 part
5	Tamra Bhasma (Ash prepared from Copper)	1 part
6	Triphala *a.* Haritaki -*Terminalia chebula* *b.* Bibhitaki- . *Terminalia belliric* *c.* Amalaki- *Emblica officinalis*	2 part
7	Shilajatu (Mineral pitch)- *Asphaltum*	3 part
8	Pura – Guggulu (gum resin)- *Commiphora mukul*	4 part
9.	Chitramool- *Plumbago zeylanica*	4 part
10.	Tikta- *Picrorhiza kurroa*	4 part
11.	Juice extract of Nimba leaf- *Azadirachta indica*	*Q.S*

The objective of the present research work was to investigate the *in-vitro* and *in-vivo* antidiabetic activity of Arogyavardhini Gutika, an ayurvedic formulation. Diabetes was induced by streptozotocin. Various parameters, like fasting plasma glucose levels, were assessed at different time intervals along with the lipid profile (total cholesterol, triglycerides, low density lipoprotein, and high density lipoprotein levels). *In-vitro* studies were performed by alpha-amylase inhibition carried

out by the starch iodine method and the dinitrosalicyclic acid method. Moreover, a pharmacokinetic study was also conducted, and the parameters Cmax, Tmax, and AUC were estimated by HPLC analysis.

MATERIALS AND METHODS

Drugs, chemicals, and reagents

The Ayurvedic formulation, Baidyanath Arogyavardhini Vati, was procured from a local market in Raipur, Chhattisgarh, India. Streptozotocin (Merk Life Science Private Limited, Mumbai), Acarbose (Healthy Life Pharma Pvt. Ltd.), Alpha-amylase (Bayer Pharmaceutical Pvt. Ltd.), 3,5 dinitrosalicylic acid (Loba Chemie Pvt. Ltd.), Soluble starch (Bayer Pharmaceutical Pvt. Ltd.), Potassium dihydrogen orthophosphate (Loba Chemie Pvt. Ltd.), Orthophosphoric acid (Loba Chemie PVT LTD Mumbai India), Sodium phosphate buffer (Loba Chemie Pvt. Ltd. Mumbai), Accu Chek active glucostrips (Roche Diagnostic India Pvt. Ltd. Mumbai), and Cholesterol Chod PAP Method Kit (Bioconcept Healthcare) were provided by the Columbia Institute of Pharmacy, Raipur, India.

In-vitro antidiabetic activity

Alpha-amylase inhibition was carried out by the starch-iodine method and the dinitrosalicyclic acid method. Alpha-amylase is responsible for the hydrolysis of alpha-bond-linked polysaccharides such as starch and glycogen into disaccharides. Hence, the inhibitory nature of the test compound reflects its antidiabetic activity because of the unavailability of glucose from GIT [10].

STARCH IODINE METHOD

The inhibition assay was performed according to standard protocol [11]. Aqueous suspension of an Ayurvedic formulation of varied concentration in 500µl was added to 500 µl of 0.02M sodium phosphate buffer (pH 6.9). 0.04 units of alpha-amylase solution were added and incubated at 37 °C for 10 minutes. A 1% starch solution was added and incubated at 37 °C for 15 minutes. 1M HCl (20 µL) was added to stop the reaction, followed by 100 µL of iodine reagent. The change in colour was noted, and absorbance was measured at 620nm. A control sample was prepared accordingly.

The % inhibition rates were calculated using formula,

$$\textbf{Inhibition (\%)} = \frac{\textbf{Abs(control)} - \textbf{Abs(extract)}}{\textbf{Abs (control)}} \textbf{ X 100}$$

DNSA METHOD [3, 5 dinitrosalicylic acid]

The dinitrosalicylic acid (DNS) method gives a rapid and simple estimation of the extent of saccharification by measuring the total amount of reducing sugars in the hydrolysate. This DNSA method tests for the presence of free carbonyl group (C=O), the so-called reducing sugars. This involves the oxidation of the aldehyde functional group present in, for example, glucose and the ketone functional group in fructose. Simultaneously, 3, 5-dinitrosalicylic acid (DNS) is reduced to 3-amino,5-nitrosalicylic acid under alkaline conditions. Because dissolved oxygen can interfere with glucose oxidation, sulfite, which itself is not necessary for the color reaction, is added in the reagent to absorb the dissolved oxygen.

The inhibition assay was performed according to standard protocol [12]. Aqueous suspension of an Ayurvedic formulation of varied concentration in 500 µl is added to 500µl of 0.02M sodium phosphate buffer (pH 6.9). 0.04 units of alpha-amylase solution

were added and incubated at 37 °C for 10 minutes. Add 500 µl of 1% starch solution to 0.02M sodium phosphate buffer (pH 6.9). The reaction will stop with the addition of 1 ml of 3, 5 DNSA reagents. The test tubes were incubated in a boiling water bath for 5 minutes and cooled to room temperature. The reaction mixture was then diluted after adding 10 ml of distilled water and measured at 540nm. A control sample was prepared accordingly.

The inhibition rates were calculated using the formula,

$$\textbf{Inhibition (\%)} = \frac{\textbf{Abs(control)} - \textbf{Abs(extract)}}{\textbf{Abs (control)}} \textbf{ X 100}$$

In-vivo antidiabetic activity

Animal

Albino Wister rats weighing 150–200 grams were used for the study. The animals were maintained under controlled conditions of temperature ($23 \pm 2\ ^0$ C) and 10-hour light and 14-hour dark cycles. The rats were randomly allocated into experimental and control groups and housed in sanitized polypropylene cages containing sterile paddy husk as bedding. The rats were provided free access to standard pellets as a basal diet and water *ad libitum*. Experiments were performed in the pharmacology laboratory of Columbia Institute of Pharmacy, Raipur (C.G.) in accordance with the CPCSEA guidelines after seeking approval from the Institutional Animal Ethics Committee (IAEC) [approval no.: CIP/IAEC/2018/116].

In-vivo antidiabetic activity (STZ) induced

Animals were fasted overnight prior to administration. Streptozotocin was administered by single intraperitoneal injection at a dose of 45 mg/kg dissolved in normal saline to induce type I diabetes [13, 14]. The diabetes was confirmed by the estimation of blood glucose levels (BGL) on the third day. Rats with BGL > 200 mg/dl were used for the study. The animals were divided into five groups, with six animals in each group.

- Group I: Normal control (vehicle treated)
- Group II: Diabetic control (vehicle treated)
- Group III: Diabetic rats treated with Arogyavardhini Gutika (200 mg/kg)
- Group IV: Diabetic rats treated with Arogyavardhini Gutika (500 mg/kg)
- Group V: Diabetic rats treated with Acarbose (500 mg/kg)

Treatment with Arogyavardhini gutika and acarbose was suspended in distilled water. The dose was given daily for 14 days to the animal by oral route, and the blood sample was collected by pricking the rat's tail for analysis of blood glucose levels. The blood samples were collected on the initial day of experiment followed by $3^{rd,}$ 7^{th}, 11^{th} and 14^{th} days of the treatment through the tail vein of rats by pricking and were immediately used for the assessment of blood glucose with a glucometer. The toxicity study of Arogyavardhini gutika has been investigated previously, and the selected dose was found to be safe [15].

Biochemical parameters [16, 17]

After the 14^{th} day of treatment, blood was collected from the retro-orbital plexus of overnight fasted rats. Blood was collected and centrifuged by using a tabletop centrifuge at 2000 rpm for 30 minutes to get serum and lipid profiles (i.e., total cholesterol, triglycerides, LDL, and HDL levels) that were estimated by the method of CHOD-PAP using standard kits, and total high density lipoprotein (HDL) was

estimated by the method of GPO-PAP using standard kits.[17] The animal experiments were conducted in a pharmacology laboratory.

Pharmacokinetic studies

The study was conducted in diabetic rats treated with Arogyavardhini gutika, and blood samples of about 0.5 mL were collected on the 11th day at predetermined time intervals from retro-orbital vein puncture using heparinized capillary tubes. Serum samples were separated after centrifugation at 8000 rpm for 15 min, and the samples were stored in a deep freezer at 20 °C until analysis by HPLC [18].

HPLC analysis

The analysis was performed by the ultra-fast liquid chromatography (Shimadzu, Kyoto, Japan) system with gradient capability and a binary pump (LC-20AD) and the analytical column C18, 250 X 4.6 mm. The column effluent was monitored with a UV-visible dual-wavelength absorbance detector (SPD-A20) at 230 nm. The mobile phase consists of methanol and 10 mM potassium dihydrogen orthophosphate (pH 3.0 adjusted with orthophosphoric acid) in the ratio 80: 20% v/v and is delivered at a flow rate of 1.0 mL/min.

Pharmacokinetic analysis

The area under the concentration versus time curve (AUC0-t) was calculated by the trapezoidal rule. The pharmacokinetic parameters, such as maximum plasma concentration (Cmax) and time of maximum concentration (Tmax), were directly obtained from the plasma concentration-time plots. Statistical significance was assessed by an unpaired Student's t-test, and the significance level of $P \leq 0.05$ was adopted for all statistical comparisons. All results are expressed as the arithmetic mean minus the standard deviation (S.D) [18, 19].

Statistical analysis

All the experiments were performed in triplicate. The data were expressed as mean [SEM]. The data were analyzed by one-way analysis of variance (ANOVA). A P - value of 0.05 was considered statistically significant. The values were calculated by Microsoft Excel. Statistical significance was assessed by an unpaired Student's t-test, and the significance level of $P \leq 0.05$ was adopted for all statistical comparisons. All results are expressed as the arithmetic mean minus the standard deviation (S.D.).

RESULTS

In-vitro study of antidiabetic activity

Arogyavardhini gutika, compared with acarbose as a standard, showed dose-dependent inhibition of alpha-amylase. The IC50 value found by the starch iodine method for Arogyavardhini gutika was 101.72 g/ml, compared with the standard carbohydrate IC50 value of 79.50 g/ml shown in figures 1(a) and 1(b). The IC50 value found by the DNSA method for Arogyavardhini gutika was 131.51 g/ml, compared with the standard Acarbose IC50 value of 82.86 g/ml shown in figures 2(a) and 2(b). Thus, the *in vitro* studies showed that Arogyavardhini gutika inhibits alpha-amylase activity by the starch-iodine method and the DNSA method. The data are shown in tables 2 and 3.

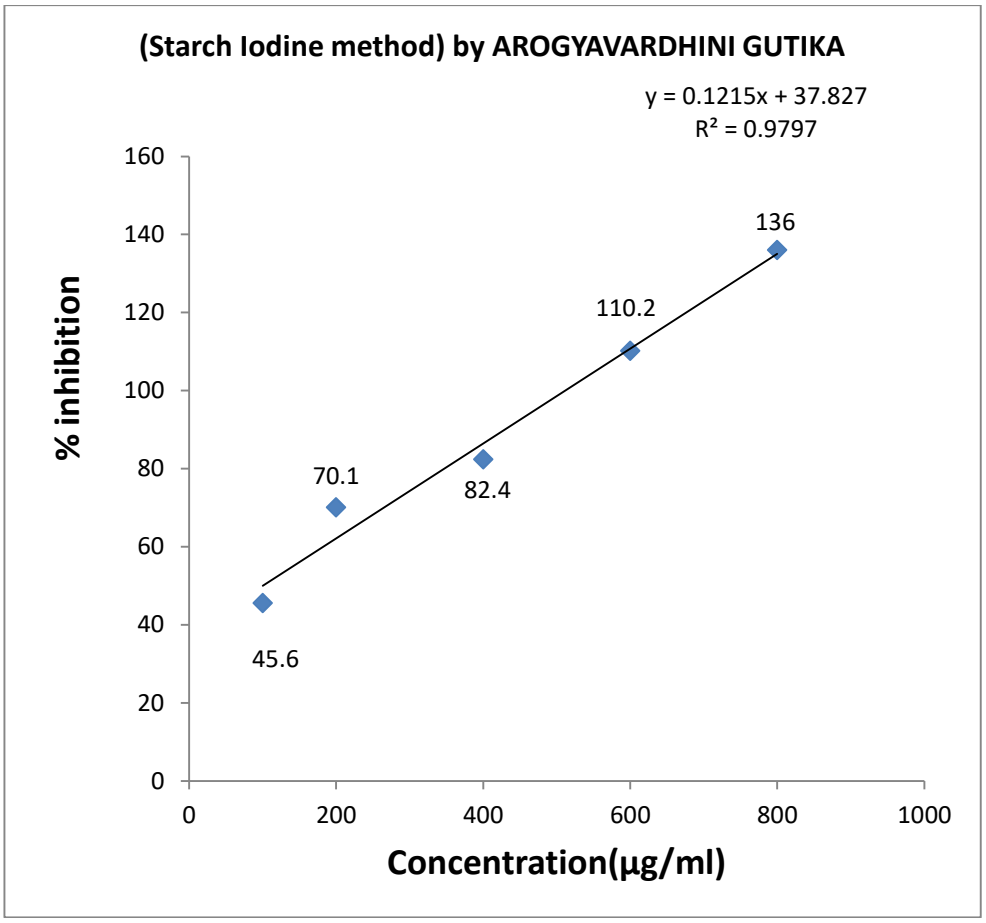


Figure 1 (A): Percentage inhibition of Alpha amylase enzyme (Starch Iodine method) by AROG-YAVARDHINI GUTIKA.

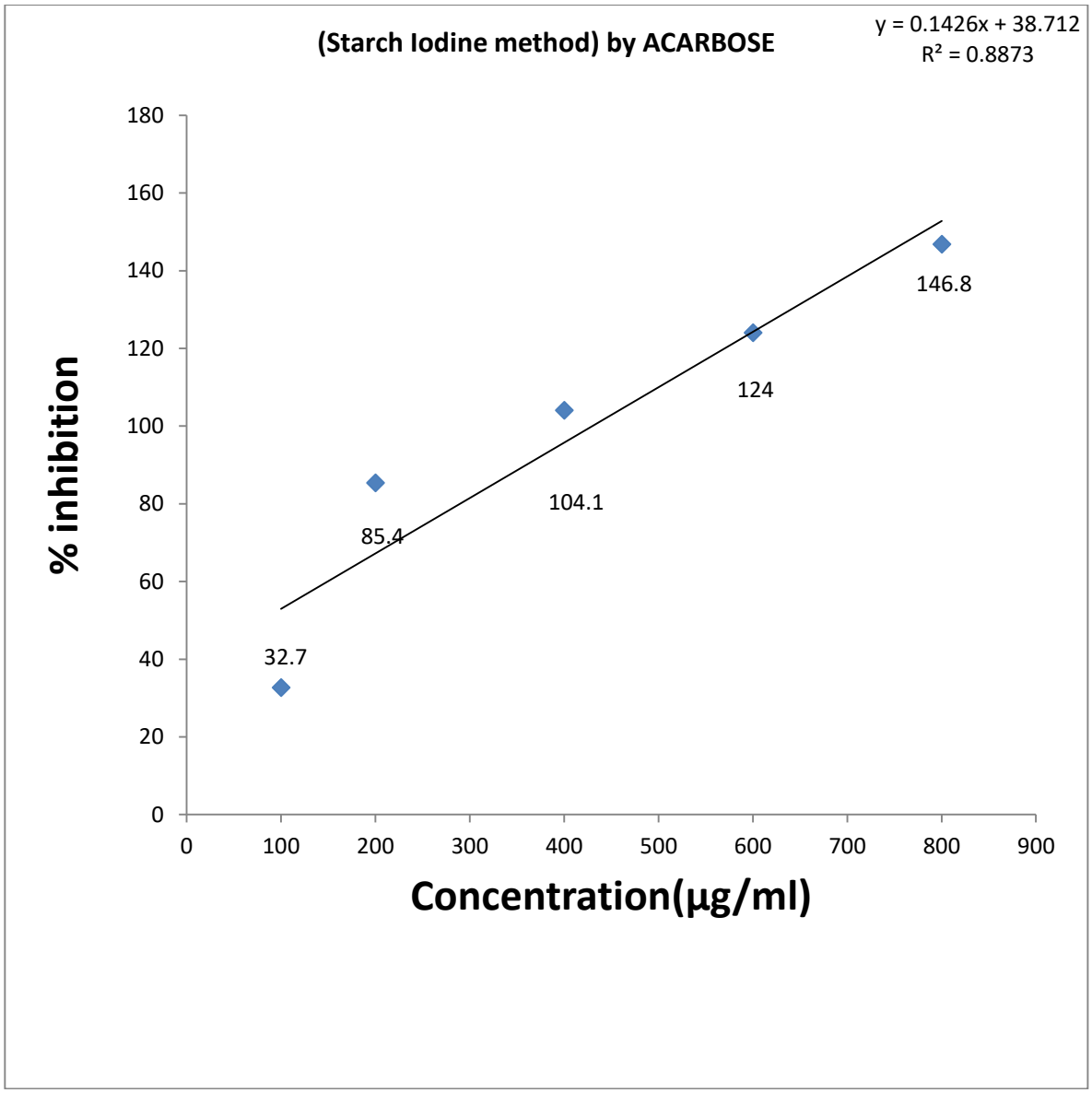


Figure 1 (B): Percentage inhibition of Alpha amylase enzyme (Starch Iodine method) by Acarbose.

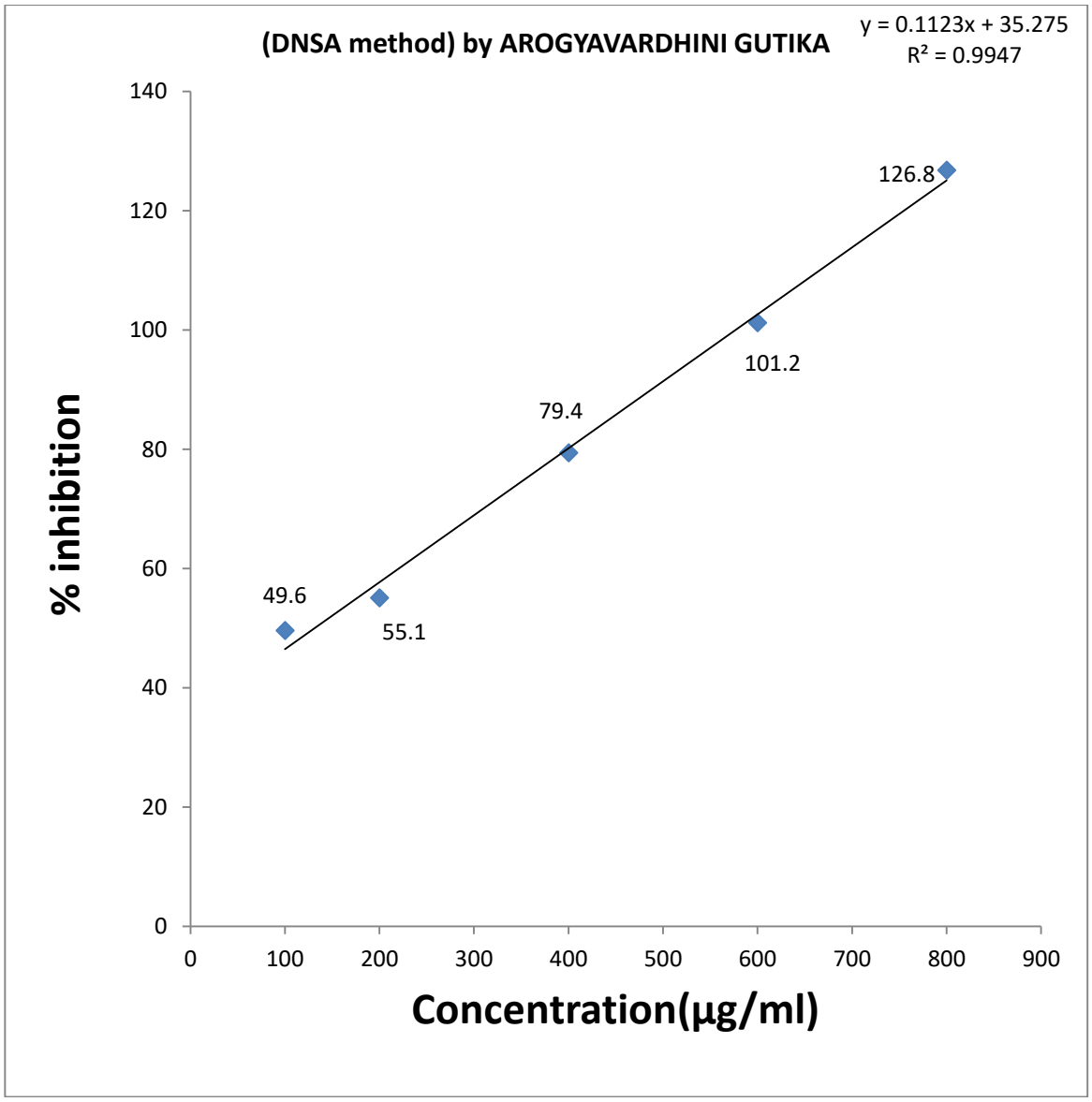


Figure 2 (A): Percentage inhibition of Alpha amylase enzyme (DNSA method) by AROGYA-VARDHINI GUTIKA.

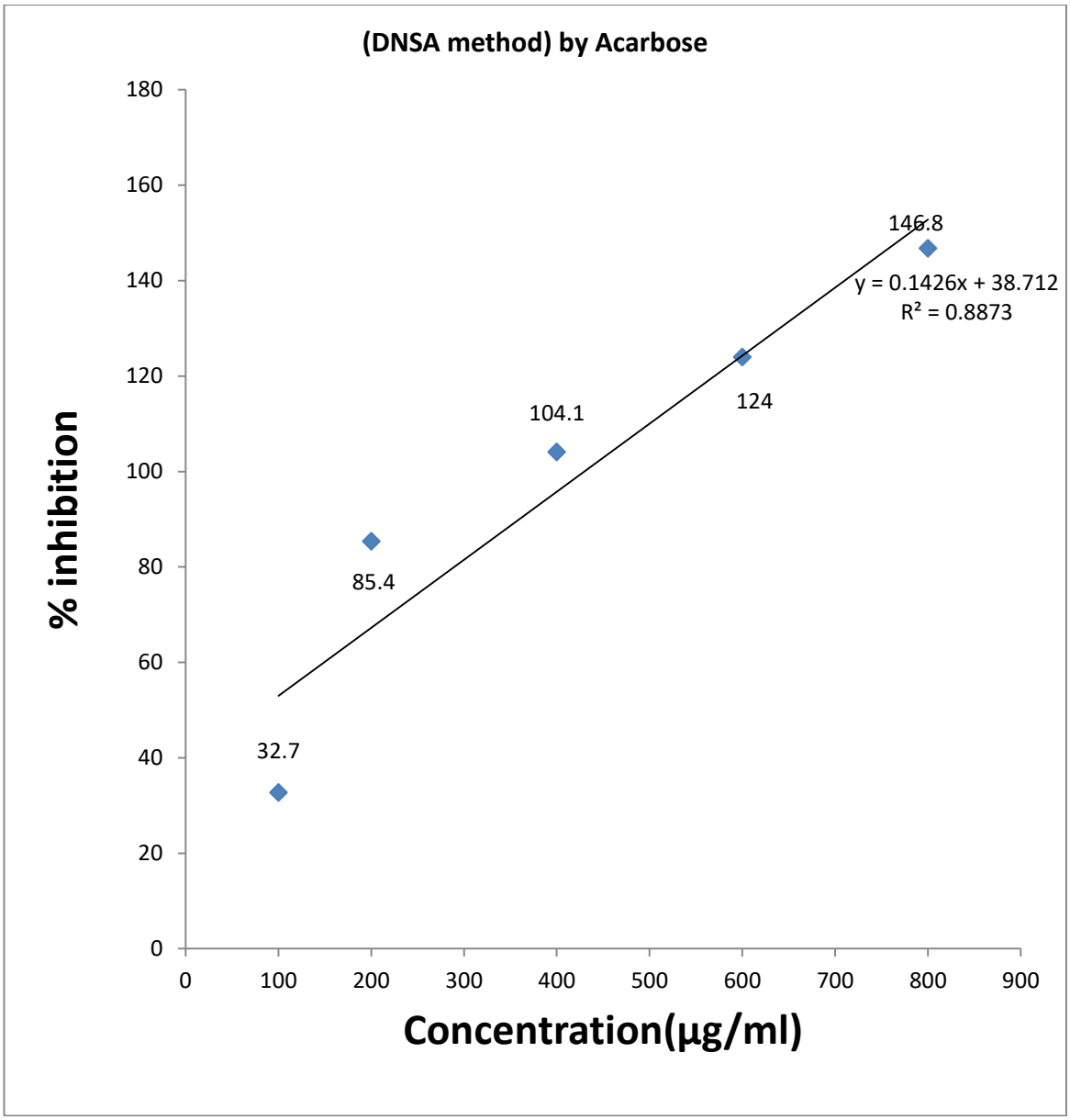


Figure 2 (B): Percentage inhibition of Alpha amylase enzyme (DNSA method) by Acarbose.

Table (2): The percent inhibition of alpha amylase (Starch Iodine method) by AROGYAVARDHINI GUTIKA *and* Acarbose at varying concentrations.

Concentration (µg/ml)	AROGYAVARDHINI GUTIKA		Acarbose	
	% inhibition	IC_{50} (µg/ml)	% inhibition	IC_{50} (µg/ml)
100	45.6±0.32	101.72	32.7±0.12	79.50
200	70.1±0.447		85.4±0.28	
400	82.4±0.447		104.1±0.21	
600	110.2±0.707		124±0.48	
800	136±0.702		146.8±0.63	

Table (3): The percent inhibition of alpha amylase (DNSA method) by AROGYAVARDHINI GUTIKA *and* Acarbose at varying concentrations.

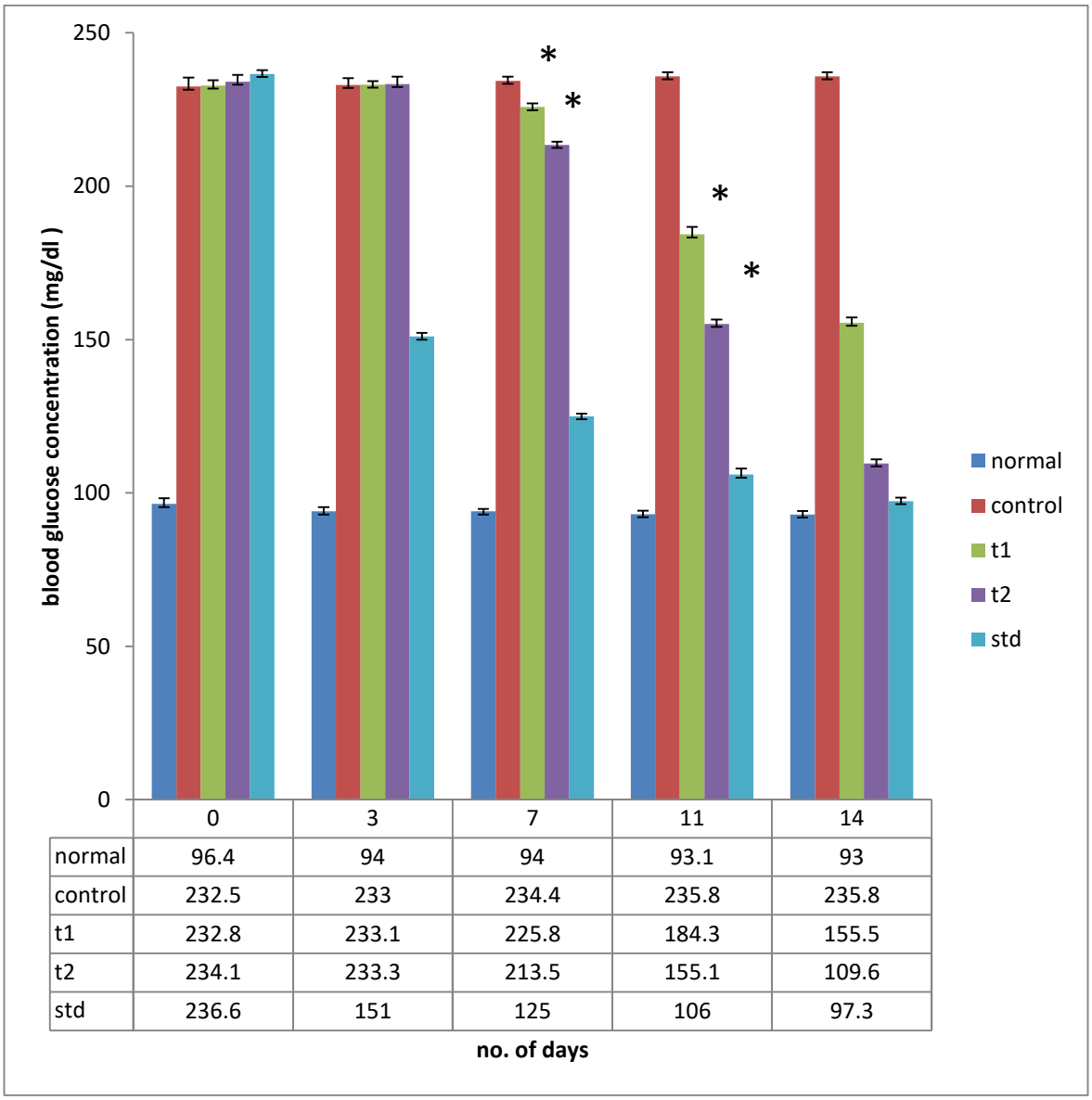
Concentration (µg/ml)	AROGYAVARDHINI GUTIKA		Acarbose	
	% inhibition	IC_{50} (µg/ml)	% inhibition	IC_{50} (µg/ml)
100	49.6±0.35	131.51	47.5±0.45	82.86
200	55.1±0.18		74.5±0.509	
400	79.4±0.53		88.4±0.678	
600	101.2±0.76		111.7±0.316	
800	126.8±0.82		146.8±0.583	

In-vivo study of anti-diabetic activity

The blood glucose level was measured in normal healthy animals before fasting and after fasting for 24 hours and induction of diabetes; again the BGL was measured on 3rd day. In normal rats; the BGL ranged from 93-96.5 mg/dl; this range conformed that rats were healthy. In case of diabetic control the rats showed BGL from 232 to 236 mg/dl from initial day to 14th day; the results confirmed that were having diabetes. However; the rats with treatment t1 receiving 200 mg/kg of Arogyavardhini gutika lowered the BGL from 233 to 155 mg/dl in overall 14 days of treatment. In last case of rats with treatment t2 receiving 500 mg/kg of Arogyavardhini gutika showed significant lowering in BGL from 233 to 109 mg/dl in overall 14 days of treatment. The results confirmed that treatment t2 receiving 500 mg/kg of Arogyavardhini gutika was best out of all the groups.

In-vivo studies indicate that raised levels of plasma glucose in STZ-induced diabetic rats were lowered by Arogyavardhini gutika at both doses (250 mg/kg and 500 mg/kg), but the effective lowering dose was 500 mg/kg. Diabetic control animals showed severe hyperglycemia compared to normal animals. It was observed that the standard drug Acarbose lowered the BGL significantly, back to a near normal level, whereas the Ayurvedic formulation at 250 mg/kg and 500 mg/kg significantly decreased the fasting blood serum glucose

level in the diabetic rats on the initial day followed by 7th and 14th days, as compared to the diabetic control group. The plasma glucose lowering activity was compared with that of acarbose, a standard antidiabetic drug, as shown in figure 3. Also in this study, the diabetic animals showed a significant decrease in body weight when compared to the control animals (Figure 4) and a change in lipid profile (Figure 5).



	0	3	7	11	14
normal	96.4	94	94	93.1	93
control	232.5	233	234.4	235.8	235.8
t1	232.8	233.1	225.8	184.3	155.5
t2	234.1	233.3	213.5	155.1	109.6
std	236.6	151	125	106	97.3

*All data presented in mean± SD (n=6), P ≤ 0.05 for test group t1 and t2

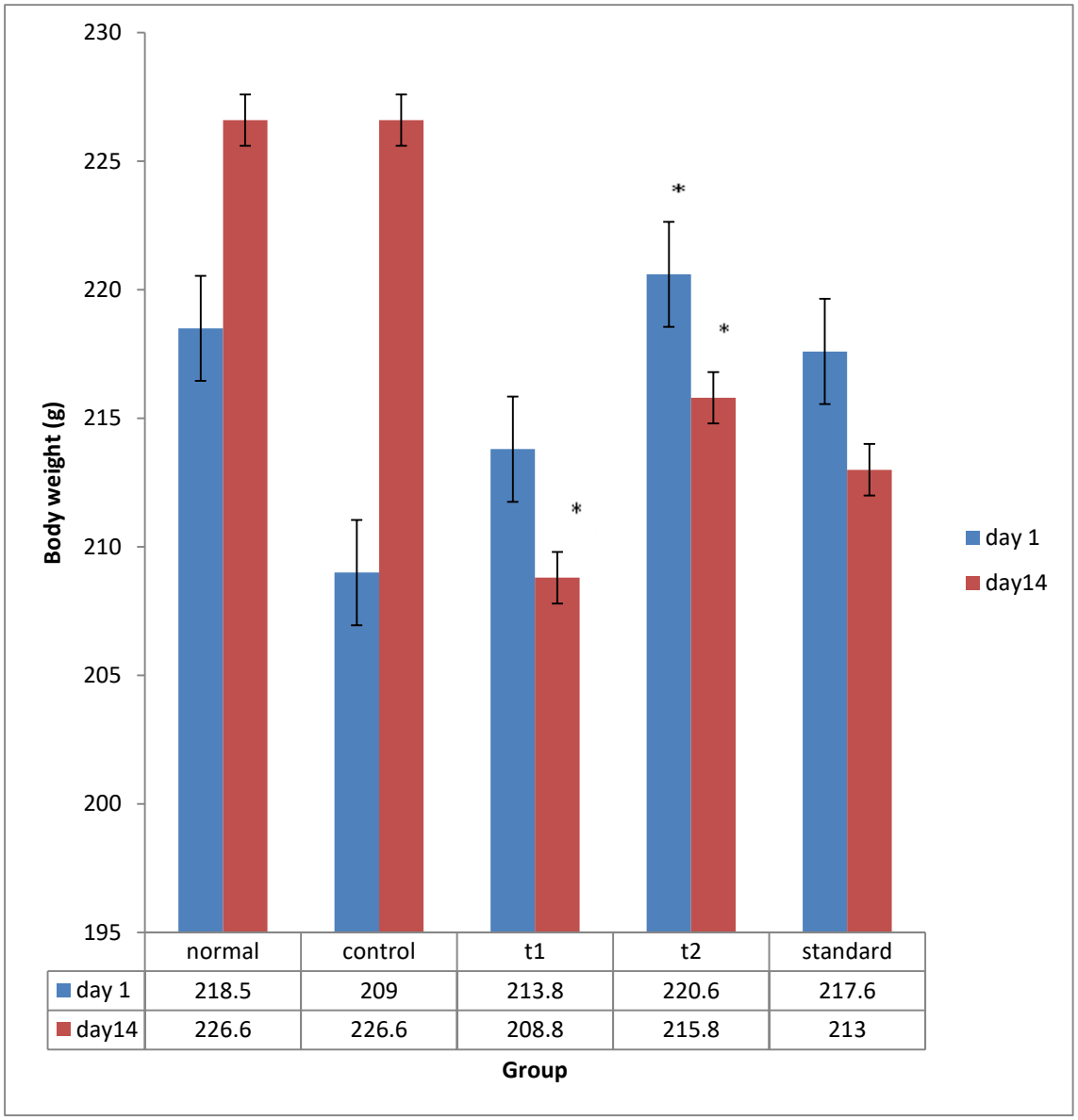
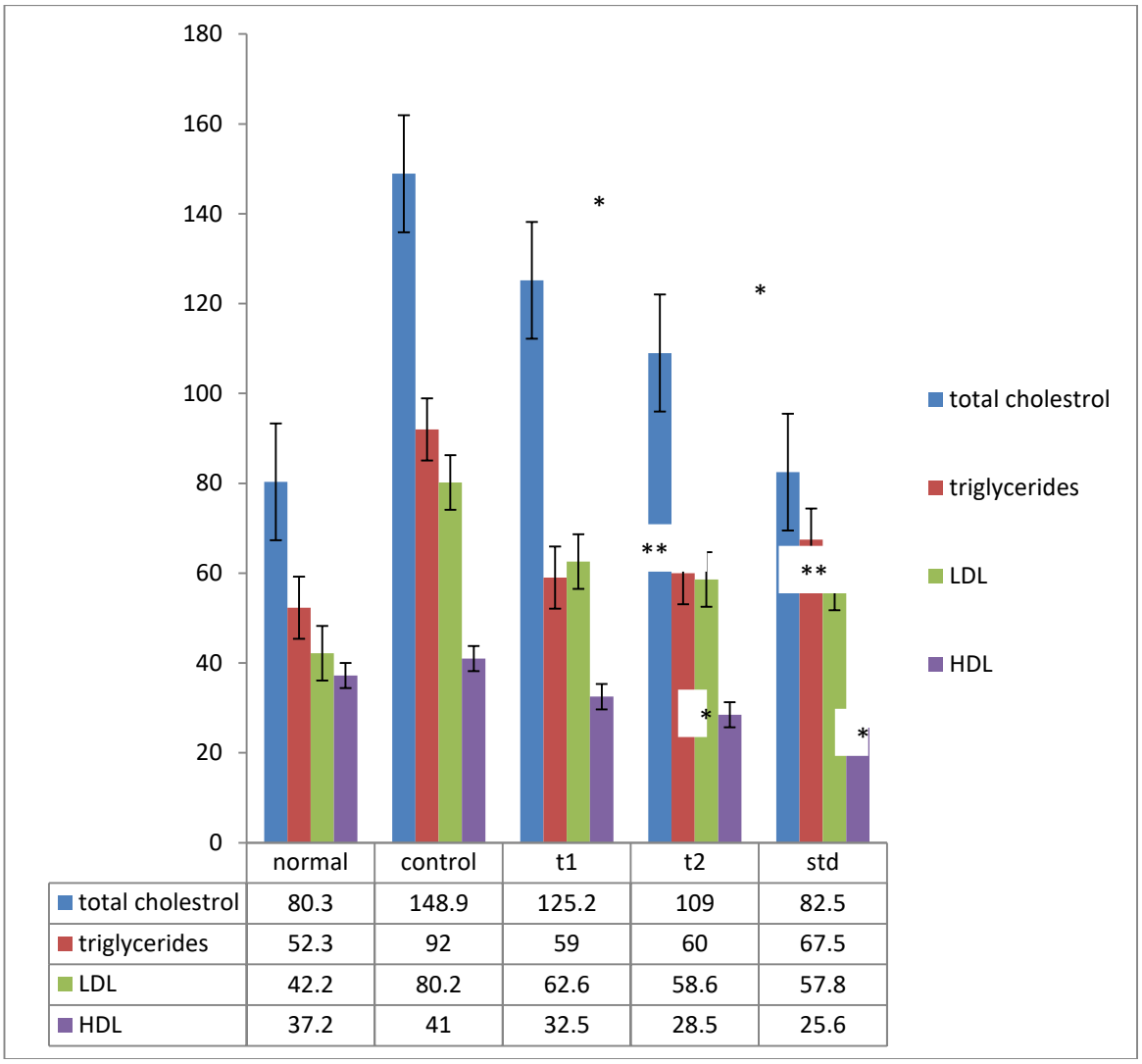


Figure (3): Effect of "Arogyavardhini gutika"in streptozotocin induced diabetic rats.

*All data presented in mean± SEM (n=6), P ≤ 0.05 for test group t1 and t2

Figure (4): Effect of "Arogyavardhini gutika" on body weight in albino rat.



	normal	control	t1	t2	std
total cholestrol	80.3	148.9	125.2	109	82.5
triglycerides	52.3	92	59	60	67.5
LDL	42.2	80.2	62.6	58.6	57.8
HDL	37.2	41	32.5	28.5	25.6

*All data presented in mean± SD (n=6), P ≤ 0.05 for test group t1 and t2

Figure (5): Effect of "Arogyavardhini gutika" on lipid profile in diabetic rats.

Relation between blood glucose level, blood lipid profile and body weight

As Body Mass Index increases, insulin resistance also increases which results in increased blood glucose level in body. Most of the studies report that high blood glucose levels are positively associated with having high total triglycerides (TG) and high total cholesterol (TC). There exists a relationship between body weight and the parameters of plasma and blood level profile with respect to age and gender. These are highlighted in our research.

Pharmacokinetic analysis

A direct relationship exists between the concentration of drug at the biophase (site of action) and the concentration of drug in plasma. Two categories of parameters can be evaluated from a plasma concentration time profile viz. Pharmacokinetic parameters and Pharmacodynamic parameters. In the present study, the pharmacokinetic parameters Cmax, Tmax, and AUC for the Arogyavardhini gutika formulation at two doses of 250 mg/kg and 500 mg/kg were estimated from plasma drug concentration profile of drugs administered at two different doses with respect to time and compared with Acarbose. The results were found to be significant at a dose of 500 mg/kg. The Tmax was the same for both doses, i.e., 2 hours, whereas the Cmax was found to be 9.75 g/ml at a dose of 250 mg/kg and 14.27 g/ml at 500 mg/kg. The data are shown in Table 4 (Figure 6).

Table (4): Pharmacokinetic parameters

PK parameters	Ayurvedic formulation (250 mg/kg)	Ayurvedic formulation (500 mg/kg)	Acarbose
Cmax (µg/ml)	9.75±0.25	14.27±0.14	24.65±0.42
Tmax (h)	2.00	2.00	2.00
AUC (µg.h/ml)	18.28±2.84	28.81±2.43	46.35±0.13

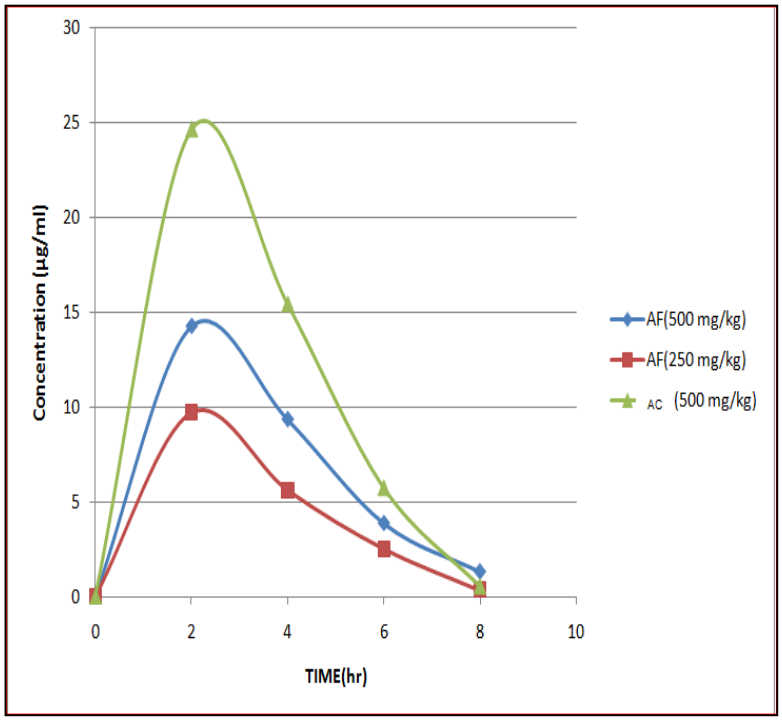


Figure (6): Mean serum concentration.

DISCUSSION

The present study was planned and conducted to evaluate the influence of Aryogravardhini gutika on blood glucose and lipid profiles in STZ-induced diabetic rats and to scientifically validate its traditional use in diabetes. The toxicity study of the formulation was as per the previous studies [15]. In order to study the influence of drugs on blood glucose in hyperglycemic conditions, drugs that slow down carbohydrate hydrolysis enzymes have been demonstrated to decrease hyperglycemia and improve impaired glucose metabolism. Therefore, the results of *in vitro* studies showed that Arogyavardhini gutika inhibits alpha-amylase activity by the starch-iodine method and the DNSA method. All through this study, the diabetic animals showed a significant reduction in body weight when compared to the control animals. It was observed that the standard drug Acarbose reduced the blood glucose level significantly, bringing it back to a near normal level, whereas the herbal formulation at 250 mg/kg and 500 mg/kg significantly ($P \leq 0.05$) decreased the fasting blood serum glucose level in the diabetic rats on the initial day of experiment followed by 3^{rd}, 7^{th}, 11^{th} and 14^{th} days as compared to the diabetic control group [19]. The present study still has certain limitations, like a lack of estimation of the pharmacokinetics of Arogyavardhini gutika. The findings of the present study suggest that Arogyavardhini vati at both doses (250 mg/kg and 500 mg/kg) showed hypoglycemia compared to normal animals, but the effective lowering dose is 500 mg/kg in type I diabetes. The Cmax and AUC of ayurvedic formulation as compared to standard carbose were low, but the Tmax of ayurvedic formulation when compared to standard was the same, i.e., 2 hours. Further, the present investigations suggest that drugs like Arogyavardhini vati may have potential effects against both type I and type II diabetes. Hence, advanced studies are needed for the assessment of chronic diabetes models.

Significance of our study

The present research work is part of a traditional system of medicine focusing on an ayurvedic formulation for the treatment of diabetes. The work is a step towards promoting the use of ayurvedic preparation for humankind's betterment of livelihood. Moreover, these preparations are easy to use and patient-friendly. The parameters studied, i.e., antidiabetic and pharmacokinetic parameters, are key parameters for the normal functioning of the human body. A pharmacokinetic study is an indication of the mechanism and extent of absorption, distribution, metabolism, and excretion of a drug administered in the body. The concentration of the drug and its half-life are important factors in drug absorption in the body. So, in our study, we investigated the antidiabetic activity through *in-vitro* and *in-vivo* methods and estimated the lipid profile parameters after administration of an ayurvedic formulation. The entire research was conducted using albino Wister rats. Experiments were done in triplicate, and the results were interpreted using software through statistical analysis.

CONCLUSION

On the basis of the present study, the *in vitro* antidiabetic activity of the herbo-mineral Aryogravardhini gutika is performed using

two methods, i.e., the starch-iodine colour assay method and the DNSA method. Arogyavardhini gutika, compared with acarbose as a standard, showed dose-dependent inhibition of alpha-amylase. Arogyavardhini gutika exhibits promising antidiabetic activity and helps maintain good glycemic and metabolic control in streptozotocin-induced rats. The antidiabetic activity of Arogyavardhini gutika at a dose of 500 mg/kg was almost close to that of the standard drug, Acarbose. Along with a remarkable reduction in TC level and body weight in STZ-induced diabetes rats. The PK parameters also revealed that the Cmax and AUC of Ayurvedic formulations as compared to standard Acarbose are low, but the Tmax of Ayurvedic formulations when compared to standard is the same, i.e., 2 hours. The Ayurvedic formulation Arogyavardhini Gutika shows potential antidiabetic activity in a dose-dependent manner and has been proven to have antidiabetic activity. It may be used as an alternative therapy for the treatment of diabetes mellitus.

ACKNOWLEDGEMENT

The authors are also grateful to the Department of Science and Technology (DST-FIST) Letter no-SR/FST/COLLEGE/2018/418, New Delhi for providing financial assistance.

REFERENCES

1] Khan A, Safdar M. Role of Diet, Nutrients, Spices and Natural Products in Diabetes Mellitus. Pakistan Journal of Nutrition, 2003; 2(1), pp. 1-12.

2] Naik SR, Mandlik RV, Desai SK. Antidiabetic activity of a polyherbal formulation. Indian Journal of Experimental Biology, 2008; 46, pp. 599-606.

3] Kaur M. Diabetes and Antidiabetic Herbal Formulations: An Alternative to Allopathy. European Journal of Internal Medicine, 2014; 6(4), pp. 226-240.

4] Pal S, Ramamurthy A, Mahajon B. Arogyavardhini Vati: A theoretical analysis. Journal of Scientific and Innovative Research, 2016; 5(6), pp. 225-227.

5] Lee HS, Yoo CB, Ku SK. Hypolipemic effect of water extracts of Picrorrhiza kurroa in high fat diet treated mouse. Fitoterapia, 2006; pp. 77:579-84.

6] Maruthappan V, Shree KS. Hypolipidemic activity of haritaki (*terminalia chebula*) in atherogenic diet induced hyperlipidemic rats. Journal of Advanced Pharmaceutical Technology & Research, 2010; 1, pp. 229-35.

7] Saravanan S, Srikumar R, Manikandan S, Jeya Parthasarathy N, SheelaDevi R. Hypolipidemic effect of triphala in experimentally inducedhypercholesteremic rats. Yakugaku Zasshi, 2007; 127, pp. 385

8] Akhtar MS, Ramzan A, Ali A, Ahmad M. Effect of Amla fruit (*Emblica officinalis* Gaertn.) on blood glucose and lipid profile of normal subjects and type 2 diabetic patients. International Journal of Food Sciences and Nutrition, 2011; 62, pp. 609-16.

9] Nohr LA, Rasmussen LB, Straand J. Resin from the mukul myrrh tree, guggul, can it be used for treating hypercholesterolemia? A randomized, controlled study. Complementary Therapies in Medicine, 2009; 17, pp. 16-22.

10] Duraisamy G, Manokaran K and Chandrasekar U. *In-vitro* α-amylase and α-glucosidase inhibitory effects of ethanolic extract of *Evolvulusalsinoides (L.).* International Research Journal of Pharmacy, 2012; 3(3), pp. 226-229.

11] Xiao Z, Storms R and Tsang A. A quantitative starch–iodine method for measuring alpha-amylase and glucoamylase activities. Analytical Biochemistry, 2006; 351, pp. 146–148.

12] Kazeem MI, Adamson JO, Ogunwande IA. Mode of inhibition of alpha-amylase and alpha-glucosidase by aqueous extract of morinda lucida benth leaf. Bio Medical Research International, 2013; 2(13), pp. 527-570.

13] Patel SS. Antihyperglycemic, antihyperlipidemic and antioxidant effect of Dihar, a polyherbal ayurvedic formulation in streptozotocin induced diabetic rats. Indian Journal of Experimental Biology, 2009; 47, pp. 564-570.

14] Singh TR, Gupta LN, Kumar N, Kumar V. Antidiabetic activity of Shilajatvadi Lauha, an Ayurvedic traditional herbomineral formulation. International Journal of Health and Allied Science, 2016; 5, pp. 9-14.

15] Kumar G, Srivastava A, Sharma SK, Gupta YK. Safety evaluation of an Ayurvedic medicine, Arogyavardhini vati on brain, liver and kidney in rats. Journal of Ethnopharmacology, 2012; 140, pp. 151–60.

16] Bangar OP, Jarald EE, Asghar S, Ahmad S. Antidiabetic Activity of a polyherbal formulation (karmin plus). International Journal of Green Pharmacy, 2009; 3, pp. 211-214.

17] Maruthappan V, Shree KS. Hypolipidemic activity of haritaki (terminalia chebula) in atherogenic diet induced hyperlipidemic rats. Journal of Advanced Pharmaceutical Technology and Research, 2010; 1(2), pp. 229-35.

18] Nagaraj and Veeresham. Effect of ashwagandha on pharmacokinetic and pharmacodynamic parameters of glimepiride in streptozotocin-induced diabetic rats. Asian Journal of Pharmaceutical and Clinical Research, 2018; 11 (4), pp. 207-210.

19] Brahmankar DM, Jaiswal SB. Pharmacokinetics: Basic Considerations In Biopharmaceutics and Pharmacokinetics A Treatise, Vallabh Prakashan, Second edition, 2009; pp. 235-39.

20] Ghule VD, Deshmukh K, Jadhav K. Conceptual Study of Arogyavardhini Vati. International Ayurvedic Medical Journal, 2021; pp. 3098-3101. doi:10.46607/iamj2809122021

21] Padmaja D, Maheshwar T, Anuradha D et.al. Arogyavardhini Vati - A Boon for Liver Disorders from Ayurveda (Fatty Liver). Ayushdhara, 2021; 8 (4), pp. 3418-3425.

22] Padmaja D, Maheshwar T, Anuradha D. Pharmaceutical Standardization and Physicochemical Analysis of Arogya-vardhini Vati. International Journal of Ayurveda and Pharma Research, 2022; 10 (8), pp. 54 – 61.