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Research Article

Anti-Diabetic Activity of Polyherbal Formulation *Aavaraiyathi churnam* in Alloxan Induced Diabetic Rats

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ABSTRACT

Diabetes mellitus is a heterogeneous disorder of carbohydrate, protein and fat metabolism in which sugars in the body are not oxidized to produce energy due to lack of pancreatic hormone insulin leads to disturbances of the acid base balance. There are two types. Type 1 (IDDM) patients have little or no ability to produce the hormone and are entirely dependent on insulin, whereas Type II (NIDDM) results from inadequate production of insulin which can be controlled by the oral hypoglycemic drugs. *Aavaraiyathi churnam* is one of the herbal based Siddha anti diabetic formulation for Type II maturity onset diabetes mellitus. The present study was aimed to evaluate the anti diabetic activity of polyherbal formulation *Aavaraiyathi churnam* in alloxan induced diabetic rats. Diabetic wistar albino rats were treated with standard drug glibenclamide and trial drug *Aavaraiyathi churnam* in two different doses 100mg and 200 mg. Hypoglycemic effect was evaluated in these rats and the efficacy of the trial drug was compared to the standard drug Glibenclamide. Oral administration of *Aavaraiyathi churnam* for 21 days in alloxan induced diabetic rats. At the end of the study period blood glucose level and body weight were statistically analyzed. Based on these results *Aavaraiyathi churnam* produced a significant reduction in blood glucose levels and increased in body weight when compared with non treated diabetic rats. So the present research work was confirmed that the polyherbal formulation *Aavaraiyathi churnam* possess hypoglycemic effect significantly.

Key Words: Diabetes mellitus, insulin, alloxan, glibenclamide, *Aavaraiyathi churnam*, blood glucose level, anti diabetic activity.

INTRODUCTION

Diabetes is one of the ancient disease. The history of diabetes is stated that in the Ebers papyrus (1500 BC).[1] Polyuria and honey urine was noted as early as 400 BC by the Indian Physician *Susrutha*. He has described this disease as '*Madhumeham'* which means- the honey in the urine [2]. Diabetes mellitus is an absolute or relative deficiency of insulin due to hyperglycemia and characterized by polyuria, polyphagia, and polydipsia. Diabetes mellitus is the commonest endocrine disorder that affects more than 171 million people worldwide and according to the resent study, this population may be increased 366 million by 2030.[3] If it is not controlled by medicine it will affect most of the internal organs such as nephropathy, neuropathy, retinopathy etc.[4] In the medical field treating diabetes mellitus without any side effects is still a challenge. In day today practice most of the diabetic patients were treated with standards anti diabetic drugs such as sulfonylureas and bigunides etc. These drugs have some kind of side effects like nausea, vomiting, abdominal pain, diarrhea, head ache etc., and thus search for a new safe and potent anti diabetic herbal formulation drug is essential to overcome these problems. As per world ethnobotanical reports, nearly 800 plants could be used to treat the diabetes mellitus. [5] In the past decade, research has been forced on scientific evaluation of traditional drugs of plant origin and screening of more effective and safe hypoglycemic agents has continued to be an important area of present day research. In developing countries 80 % of population is using traditional medicine in primary medical problems.[6] In Siddha system of Medicine, many single and poyherbal formulations and higher medicines like *Parpam, Chendooram* and *Chunnam* have been practiced to cure or control diabetes mellitus from time immemorial. One of the well known anti diabetic drug in Siddha system of medicine is *Aavaraiyathi churnam* comprising of five indigenous medicinal plants namely *Cassia auriculata* leaves, flower, seed, bark and root bark, *Odina wodier* bark, *Coscinium fenestratum* stem, *Ficus glomerata* tender leaves and *Cocculus cordifolius* stem.[7] All these plants are well known in the Siddha system of medicine from ancient time for their various therapeutic properties especially hypoglycemic activity.[8-12] But this formulation is not yet evaluated scientifically. Hence an attempt has been made to determine the anti diabetic potential of *Aavaraiyathi churnam* on the serum blood glucose level in alloxan induced diabetic rats.

MATERIAL AND METHODS

Plant Materials*: Cassia auriculata* (*Aavarai*) leaves, flower, seed, bark and root bark each 35 gm, *Odina wodier* (*Odhiyam*) bark 87.5 gm, *Coscinium fenestratum*

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Table No 1: Effect of *Aavaraiyathi churnam* on body weight in alloxan induced diabetic animal.

Body weight in 21 days (gm) (Mean±SD)					
Groups n=(6)	Initial	1 day	7 days	14 days	21 days
Group I	183.16±3.48	183.16±3.48	178.33±4.76	175.00±5.47	171.33±4.92
Group II	182.00±5.17	171.33±5.71	176.50±7.14	180.16±6.04***	184.16±5.45***
Group III	183.66±3.93	171.66±3.32	174.33±3.50	177.66±4.03	182.33±4.17**
Group IV	184.16±4.02	171.66±4.17	176.66±4.17	181.50±4.23***	187.00±4.00***

*Values are MEAN ± SD (n=6) one way ANOVA followed by Dunnet's multiple comparison test. Where the values are *** P<0.001; ** P<0.01 when compared with control group.*

Table No 2: Anti diabetic activity of *Aavaraiyathi churnam* extracts against alloxan induced diabetic rats.

Blood sugar level in 21 days mg/dl (mean ± SD)					
Groups (n=6)	Initial	1 day	7 day	14 day	21 day
Group I	255.50±8.06	263.66±6.94	276.66±6.53	283.66±6.88	297.66±4.13
Group II	255.00±12.49	157.00±7.69	117.66±10.96	103.66±3.88	93.00±5.47***
Group III	251.66±8.26	213.00±13.76	174.50±6.92	144.83±5.60	124.33±5.98***
Group IV	254.16±8.20	197.66±5.57	154.66±2.42	123.33±2.42	103.33±2.06***

*Values are MEAN ± SD (n=6) one way ANOVA followed by Dunnet's multiple comparison test. Where the values are *** P<0.001; ** P<0.01 when compared with control group.*

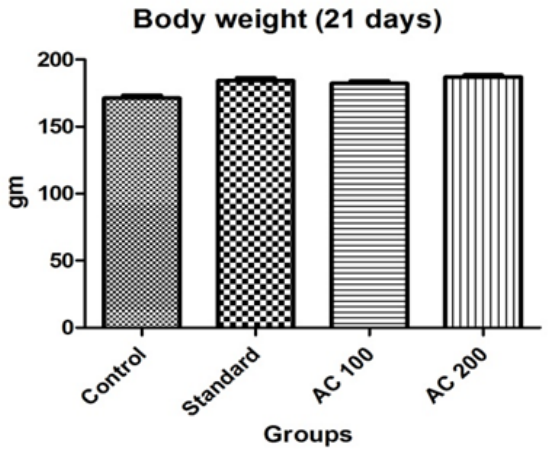

Fig No.1. Showing the diagrammatic approach of body weight of the animal groups.

(*Maramanjal*) stem 43.75, *Ficus glomerata* (*Atthi*) tender leaves 21.75 gm, *Cocculus cordifolius* (*Seenthil*) stem 183 gm were collected from in and around Chennai. All the plants were identified and authenticated by the botanist, Government Siddha Medical College, Chennai. The vouchers of specimen samples of the plants were kept in the department for future reference. After identification the above plants were cleaned well with water and dried in a shadow place. After complete drying, they powdered separately and mixed all together and the trial medicine was prepared as per the classical text. Good manufacturing practice was emphasized during the preparation. Then the prepared drug is kept in air tight container which was used for this study.

Animals: Healthy Wistar albino rats of either sex and the age of six to eight weeks weighing 160-200 g were used for this study. Before starting the experiment, the animals were acclimatized to the laboratory conditions for a period of 2 weeks at an ambient temperature (24±2 ºC) and relative humidity (40-60%). The light - dark cycle was followed. [13] The animals were fed with standard

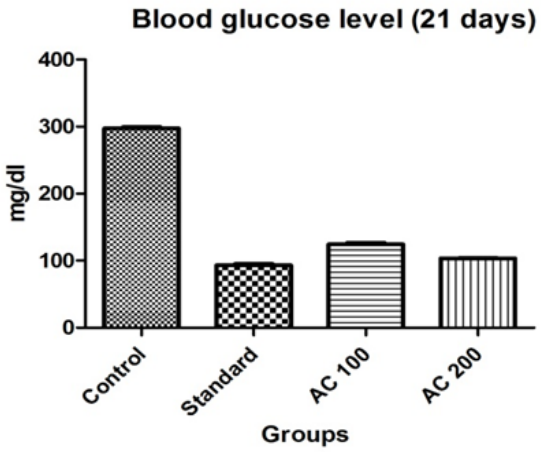

Fig No.2. Showing the diagrammatic approach of blood glucose level of the animal groups.

laboratory diet and water *ad libitum*. The animals were fasted for overnight before the study but had free approach to water. All the experimental procedure and protocols were reviewed and approved by the Institutional Animal Ethical Committee (IAEC) and all the experiments were carried out by following the guidelines of CPCSEA.

Induction of diabetes with alloxan: Diabetes was induced in Wistar albino rats by using a single intra peritoneal injection of alloxan monohydrate (150mg/kg) dissolved in normal saline. One hour after administration of alloxan, the animals were fed with standard laboratory diet and water *ad libitum*.[14] Three days after alloxan injection, the blood samples were collected from tail vein using capillary tubes and blood glucose level was analyzed.[15] Blood glucose level range > 200 mg/dl were included for further study.

Experimental Design: The selected diabetic rats were divided randomly into four groups with six animals in each group.

Group 1: Served as diabetic control group.

Group 2: Diabetic rats treated with standard drug glibenclamide (10 mg/ kg) orally for 21 days.

Group 3: Diabetic rats given *Aavaraiyathi churnam* (100 mg/kg) [AC 100] orally for 21 days.

Group 4: Diabetic rats given *Aavaraiyathi churnam* (200 mg/kg) [AC 200] orally for 21 days.

The administration of the trial drug and standard drugs were carried out every day for 21days. Blood samples were collected through the tail vein just prior to and on days 1,7,14 and 21 after the drug administration and the percentage of reduction in blood glucose was estimated by using glucometer and compared.

STATISTICAL ANALYSIS

The results were represented as Mean ± Standard Error Mean. The statistical significance was computed using One Way ANOVA followed by Dunnet's multiple comparison test and compared with diabetic untreated control group with Standard, AC 100 and AC 200 where the n=6 animals in each one group were used. $P<0.05$ was considered statistically significant.

RESULTS AND DISCUSSION

One of the most commonly used chemical agent predominantly in laboratories to induce diabetes in animal is alloxan which is an oxidized product of uric acid that tends to destroy the islet cells of the pancreas by oxidation mechanism and producing Type 1 diabetes known as '*alloxan diabetes'*. The present study was evaluated the anti diabetic activity of *Aavaraiyathi churnam* against alloxan induced diabetic rats. The continuous treatment of the *Aavaraiyathi churnam* for the period of 21 days in two different doses (100 mg/kg and 200 mg/kg of body weight). Glibenclamide is the standard drug is used to stimulate insulin from β cells of islets of langerhans for many years. Hence Glibenclamide (10mg/Kg) was selected as standard drug.

The results of the body weight and blood glucose level of diabetic control group, standard group (Glibenclamide 10 mg/kg) and two different doses of trial drug *Aavaraiyathi churnam* (100mg and 200mg/kg b.w) [AC 100 and AC 200] were summarized in Table No 1 and Table No 2 respectively. Data are statistically obtained by paired't' test to analyze the significance of standard and trial drug between the test and control groups.

In Table No.1, the body weight of the diabetic untreated control group was decreased from 183.16±3.48 to 171.33±4.92 after 21 days. The standard drug group the initial body weight was 171.33±5.71 and after three weeks of treatment was 184.16±5.45. The initial body weight of AC 100 and AC 200 were 171.66±3.32, 171.66±4.17 respectively and after 21 days of treatment the bodyweight was gained to 182.33±4.17 and 187.00±4.00 respectively, which are highly and extremely significant when compared with control group. When compared between the two different doses of trial drug [AC100 and AC200], AC200 gained more weight than the AC100 group. Two different doses of the trial drug *Aavaraiyathi churnam* (AC 100 and AC 200) blood glucose level were studied in two different groups of animals. Both groups showed a extremely significant decrease of blood glucose level on alloxan induced diabetic rats when compared to control group. The initial readings of blood glucose level of AC100 and AC200 were 251.66±8.26 and 254.16±8.20 respectively. After

the trial period both doses produced consistent reduction in the blood glucose levels after 7 days (174.50±6.92, 154.66±2.42) and marked reduction in 21 days (124.33±5.98 and 103.33±2.06). However AC 200 has shown maximum effect than AC100. In standard group initial blood glucose level was 255.00±12.49 and the post test was 93.00±5.47 which showed that the standard drug produced maximum hypoglycemic effect and the statistical analysis was extremely significant and slightly higher than that of trial drug groups. Untreated diabetic rat group showed increase in blood glucose level throughout the entire study period. Initially blood glucose level of untreated diabetic control group was 255.50±8.06 and after 21 days of trial period the blood glucose level was increased to 297.66±4.13. The results were summarized in Table No.2.

Based on the results it can be observed that there was an extremely significant reduction in blood glucose level by trial drug *Aavaraiyathi churnam* in alloxan induced diabetic rats. The maximum result obtained from AC 200 mg dose level which showed the dose dependent anti diabetic activity of the trial drug *Aavaraiyathi churnam*. The declined trend was observed at a constant level. The anti diabetic activity of *Aavaraiyathi churnam* may be due to the increased release of insulin from beta cells of pancreas or it may potentiate the effect of insulin. Treatment of *Aavaraiyathi churnam* in diabetic rat also showed the highly significant weight gain property which favours the beneficial effect of *Aavaraiyathi churnam* in treating diabetic patients successfully.

CONCLUSION

Aavaraiyathi churnam is a combination of five herbs and it is more effective in the treatment of diabetes mellitus as determined by extreme statistically significant p-value < 0.0001 in alloxan induced diabetic rats. The mechanism of the hypoglycemic activity of poly herbal formulation *Aavaraiyathi churnam* may be due to enhance the effect of insulin and stimulates the insulin secretion from beta cells of pancreas. So this study suggests that the *Aavaraiyathi churnam* had potent anti diabetic effect which could be used in the management of diabetes mellitus effectively.

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