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A Study of Prophylactic Effect Against Diabetes of Two Ayurvedic Drugs 'Jambadyarista' and 'Bohumutrantak Ras' in Normal as well as Alloxan-induced Diabetic Rats

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Authors' contributions

This work was carried out in collaboration between all authors. Authors MSA and AAC designed and wrote the research protocol. Authors TUI, ASB, MRS and TS performed the experiments, managed the literature searches. Authors ZS, AR and AH helped to the experimentations and performed the statistical analysis. Author MSA has taken care of the whole project during the research period. All authors read and approved the final manuscript.

Original Research Article

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ABSTRACT

Aims: The present study was designed to evaluate the anti-diabetic as well as prophylactic activity of Jambadyarista and Bohumutrantak Ras in alloxan-induced diabetic rats.
Study Design: Study the prophylactic and antiglycemic effects against diabetes of two Ayurvedic drugs 'Jambadyarista' and 'Bohumutrantak Ras' in normal as well as alloxan-induced diabetic rats using *in vivo* models.

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Place and Duration of Study: Department of Pharmaceutical Chemistry, Faculty of Pharmacy, University of Dhaka, Dhaka-1000, Bangladesh between December 2013 and March 2014.
Methodology: To investigate the activity, 70 Long Evans rats divided into seven (A-G) groups were included in this study. Diabetes was induced by single intraperitoneal injection of alloxan (150mg/kg body weight) while the Ayurvedic drugs Jambadyarista and Bohumutrantak Ras were given orally at a dose of 200mg/kg of body weight. Group A was control group. Groups B and E were allowed to fast for 12 hours. Diabetes was induced by intraperitoneal injection of freshly prepared solution of alloxan (150mg/kg) in normal saline after base line glucose level determination.
The alloxan-treated rats were allowed to food over night to overcome drug induced hyperglycemia. After 48 hours, blood glucose level was measured with an Accu-Chek glucometer using blood sample from the tail vein of each rat. Diabetes was established in animals when blood glucose level was raised to 11.1-32.6mmol/L. After the establishment of diabetes, the experiments were carried out.
Groups D and G were allowed to induce diabetes by single intraperitoneal injection of alloxan (75mg/kg body weight) in normal saline every day for 5 days.
Results: In case of alloxan-induced diabetic rats, Jambadyarista and Bohumutrantak Ras showed prophylactic activity against diabetes as well as also reduced blood glucose level. By statistical analysis of results, it was found that Jambadyarista and Bohumutrantak Ras have prophylactic activity against diabetes in normal and alloxan- induced diabetic rats.
Conclusion: It can be inferred that the Ayurvedic drug “Jambadyarista and Bohumutrantak Ras” significantly possess prophylactic activity against diabetes. They also showed antidiabetic effect in alloxan-induced diabetic rats. Further comprehensive cellular and molecular investigations are required to characterize the exact mechanism responsible for its prophylactic and antidiabetic effects.

Keywords: Diabetic; prophylaxis; alloxan; jambadyarista; bohumutrantak ras.

1. INTRODUCTION

In the last few years, there has been an exponential growth in the use of herbal medicine and these drugs are gaining popularity both in developing and developed countries because of their natural origin and fewer side effects [1]. Many traditional medicines in use are derived from medicinal plants, minerals and organic compounds [2]. The use of plant extracts is extremely popular in large numbers of the world’s population, particularly in Asia and Europe [3]. However, for medicine as practiced in the Western countries, one observation that appears to be forgotten is that many of the pharmaceutical agents currently prescribed appear to have been derived from natural compounds found in traditional medicinal plants [4]. As a specific example, metformin of biguanide group is considered one of the first-line agents used for the treatment of type 2 diabetes, and its use can be traced to the traditional use of *Galega officinalis* to treat diabetes and the subsequent search to identify active principle with reduced toxicity [5]. However, it has been reported that more than 1200 traditional plants may have been used for real or perceived benefit of medicinal purposes for the treatment of diabetes [6,7]. Among them extract of seed of *Syzyzium cumini*, known as black plum, Bengali name "Kalo Jam" is widely used in Bangladeshi folk medicine for the treatment of diabetes mellitus [8]. Jambadyarista is a liquid oral preparation of extract of fruit and seed of *S. cumini*. The leave, bark and seeds are the most useful parts among which the seeds are popular for their antidiabetic property. The major ingredient of

the plant that shows antidiabetic action is jamboline in the seeds. The unique phenomenon in black plum seeds is that the main ingredient jamboline would not exhibit antidiabetic property alone. With the catalytic activity and association with other inorganic or inactive substances known as intraplant synergy in the seed, it exhibits the antidiabetic activity. In traditional Ayurvedic medicine, black plum is also used to help with thirst, diarrhea and in healing of wounds [9]. *S. cumini* also contains a range of essential oils and different alkaloids which are responsible for antibacterial activities [10,11]. The Ayurvedic preparation "Bohumutrantak Ras" consists of different classes of natural ingredients such as Rasa-Sindoor, *Opium poppy, Ficus carica, Aegle marmelose, Piper cubeba.*

The present study was designed to evaluate the prophylactic activity against diabetes of Jambadyarista and Bohumutrantak Ras in normal and alloxan-induced diabetic rats.

2. MATERIALS AND METHODS

2.1 Drugs, Reagents and Chemicals

Alloxan (Merck, Germany) was purchased from the local market, and Jambadyarista and Bohumutrantak Ras were purchased from Shadhana Aushadalaya Ltd., Dhaka, Bangladesh and Modern Herbal Research Garden, Demra, Dhaka, Bangladesh, respectively. All other reagents such as methanol, ethanol were of analytical grade and purchased from Active Fine Chemicals Ltd., Bangladesh.

2.2 Instruments

Glucometer and Kits (Accu-Chek Active, Germany).

2.3 Experimental Animals

A total of 70 Long Evans rats of either sex weighing about 150-200g, aged 2 months were purchased from animal house of the Department of Pharmacy, Jahangirnagar University, Savar, Dhaka, Bangladesh. Prior to commencement of the experiment, all the rats were acclimatized to the new environment for a period of one week. During the experiment period the rats were kept in a well ventilated animal house at room temperature of 25ºC. They were supplied with standard pellets and fresh drinking water. All the rats were kept in cage and maintained with natural 12hr light and dark cycle in the Animal House of the Institute of Food and Nutrition Science, University of Dhaka, Bangladesh. This study was conducted according to the Declaration of Helsinki.

2.4 Experimental Rat Grouping

70 Long Evans rats of both sexes were randomly assigned into seven groups (Group A-G). 10 rats were taken in each group.

2.5 Experimental Induction of Diabetes

Groups B and E were allowed to fast for 12 hours. Diabetes was induced by intraperitoneal injection of freshly prepared solution of alloxan (150mg/kg) in normal saline after base line glucose level determination.

The alloxan-treated rats were allowed to food overnight to overcome drug induced hyperglycemia. After 48 hours, blood glucose content was measured with an Accu-Chek glucometer using blood sample from the tail vein of the rats. Diabetes was established in animals when blood glucose level was raised to 11.1-32.6mmol/L. After the establishment of diabetes, the experiments were carried out.

Groups D and G were allowed to induce diabetes by single intraperitoneal injection of alloxan (75mg/kg body weight) in normal saline every day for 5 days.

Group A: This group was marked as control group. After each 7 days, blood glucose was measured by Accu-Chek Active glucometer till the experiment was completed. This group was provided with only normal water and pellets as food supplement during the experiment.

Group B: In this group, diabetes was induced by single intraperitoneal injection of alloxan (150mg/kg body weight) for 3 days. Then the rats were treated with Jambadyarista everyday for a total of 14 days to evaluate antidiabetic effect.

Group C: In this group, Jambadyarista was given orally at a dosage of 200mg/kg of body weight for 30 days for prophylactic study and induction of production of the antibody. During the treatment with the drug, alloxan (150mg/kg body weight) was injected from day 21 to day 30 by intraperitoneal route to evaluate the prophylactic effect.

Group D: In this group, diabetes was induced by single intraperitoneal injection of alloxan (75mg/kg body weight) followed by treatment with Jambadyarista on the same day every day for 5 days. At everyday afternoon, the blood glucose level was measured.

Group E: In this group, diabetes was induced by single intraperitoneal injection of alloxan (150mg/kg body weight) for 3 days. Then the rats were treated with Bohumutrantak Ras everyday for a total of 14 days to evaluate antidiabetic effect.

Group F: In this group, Bohumutrantak Ras was given orally at a dosage of 200mg/kg of body weight for 30 days for prophylactic study and induction of production of the antibody. During the treatment with the drug, alloxan (150mg/kg body weight) was injected from day 21 to day 30 by intraperitoneal route to evaluate the prophylactic effect.

Group G: In this group, diabetes was induced by single intraperitoneal injection of alloxan (75mg/kg body weight) followed by treatment with Bohumutrantak Ras on the same day every day for 5 consecutive days. At everyday afternoon, the blood glucose level was measured.

2.6 Statistical Analysis

The results were expressed as mean±standard deviation. The significance of the results were analyzed by '*t*' tests. A '*p*' value of 0.05 was taken as significant. All statistical analyses were performed using Kaleida Graph (Synergy Software, 2457 Perkiomen Avenue, Reading, PA 19606-2049 USA).

3. RESULTS

In the present study, diabetes was induced in rats by injecting alloxan (150mg/kg body weight) intraperitoneally. Alloxan is a cytotoxic agent for inducing diabetes in a wide variety

of animal species by damaging insulin secreting pancreatic β-cells, resulting a decrease in endogenous insulin release, which paves the way for the decreased utilization of glucose by the tissues. In case of alloxan-induced diabetic rats (Group B and E), Jambadyarista and Bohumutrantak Ras reduced blood glucose level significantly (Figs. 1 and 2).

This work has also evaluated the prophylactic effect of the Ayurvedic drug Jambadyarista and Bohumutrantak Ras on fasting blood glucose level in normal as well as alloxan-induced diabetic rats. In groups C and F, the rats were treated with Jambadyarista and Bohumutrantak Ras continuously up to 30 days; from 21st day of the treatment, alloxan (150mg/kg) was injected to the rats for 7 consecutive days. There was no significant change of blood glucose level after introduction of alloxan which indicated that Jambadyarista and Bohumutrantak Ras prevented hyperglycemia of the rats by alloxan (Figs. 3 and 4).

The prophylactic effect of the Ayurvedic drug Jambadyarista and Bohumutrantak Ras has also been evaluated in alloxan-induced diabetic rats. In groups D and G rats, diabetes was induced by single intraperitoneal injection of alloxan (75mg/kg body weight) followed by treatment with Jambadyarista every day for 5 consecutive days. At everyday afternoon, the blood glucose level was measured. During the experiment, the blood glucose levels were found almost unchanged i.e. there was no significant change of blood glucose level after introduction of alloxan which indicated that Jambadyarista and Bohumutrantak Ras prevented hyperglycemia of the rats by alloxan (Figs. 5 and 6).

Experiments on male and female rats showed that Jambadyarista and Bohumutrantak Ras have prophylactic effect against diabetes as well as antihypoglycemic property, which suggest its potentiality to induce immunity against type 1 diabetes mellitus, irrespective of gender of animals.

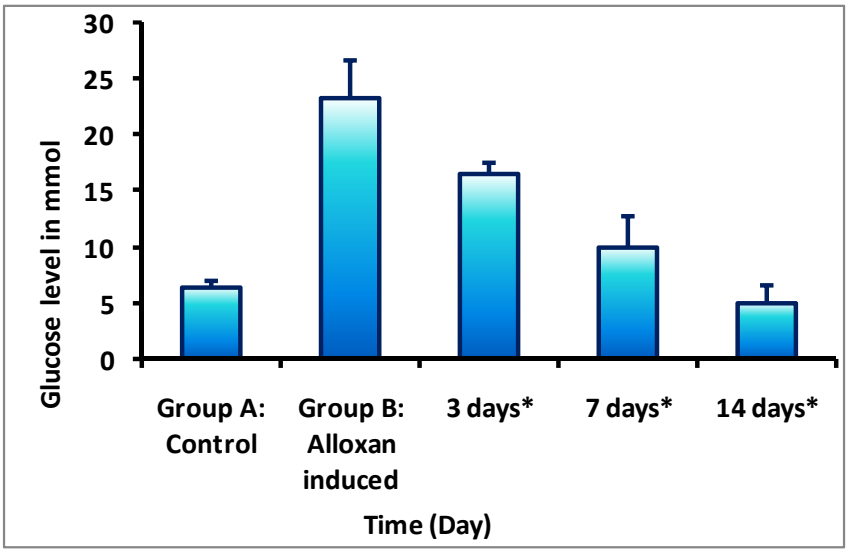

Fig. 1. Blood glucose level after giving ayurvedic drug jambadyarista in alloxan-induced rats. The standard deviations are shown as error bars. The differences in the curves are statistically significant (p=0.05) which have been shown by asterisks

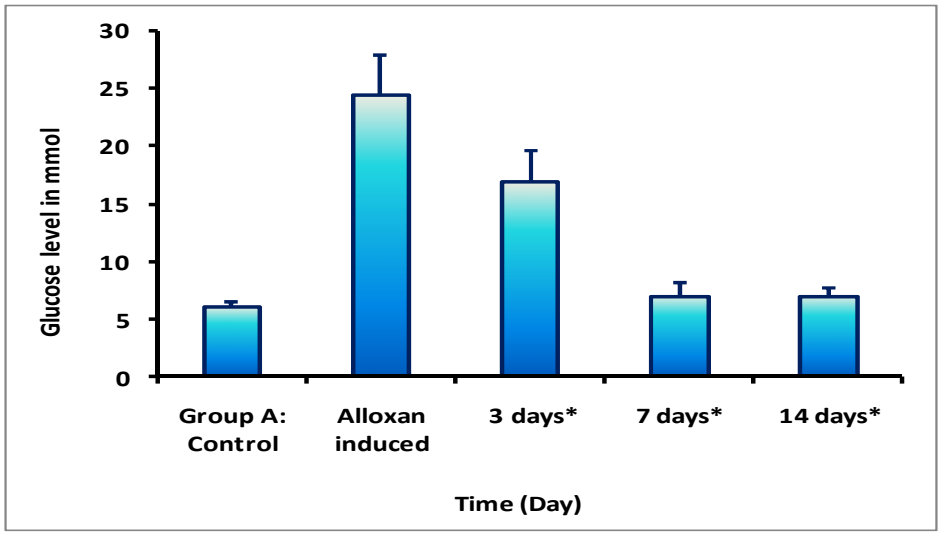

Fig. 2. Blood glucose level after giving ayurvedic drug bohumutrantak ras in alloxan-induced rats. The standard deviations are shown as error bars. The differences in the curves are statistically significant (*p*=0.05) which have been shown by asterisks

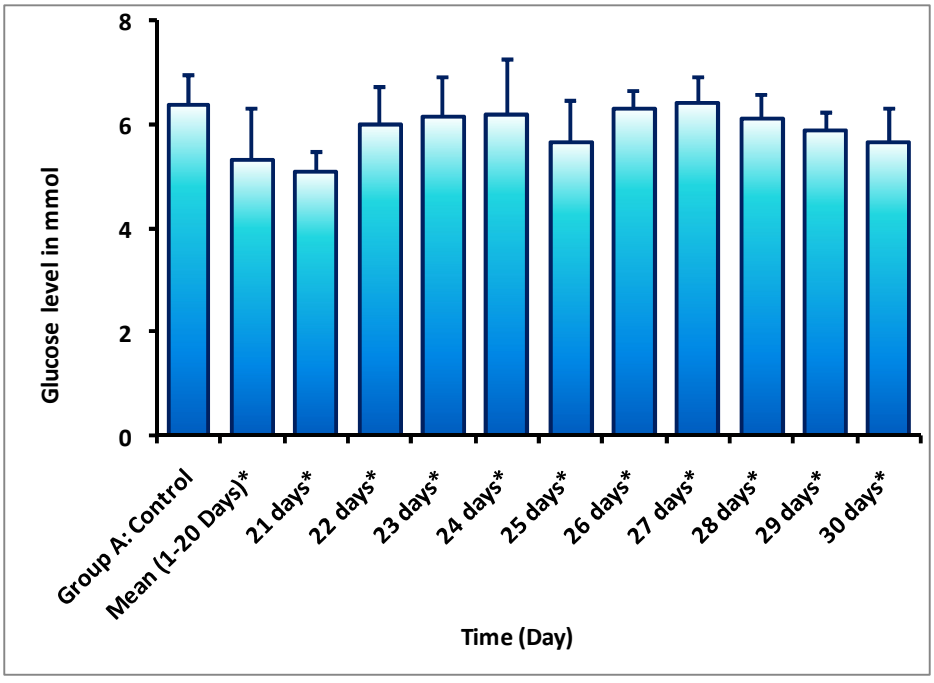

Fig. 3. Blood glucose level for prophylactic study after giving ayurvedic drug Jambadyarista in rats at dosage of 200mg/kg of body weight for 30 days followed by alloxan (150mg/kg body weight) from 21 to 30 days by intraperitoneal route. The standard deviations are shown as error bars. The differences in the curves are statistically significant (*p*=0.05) which have been shown by asterisks

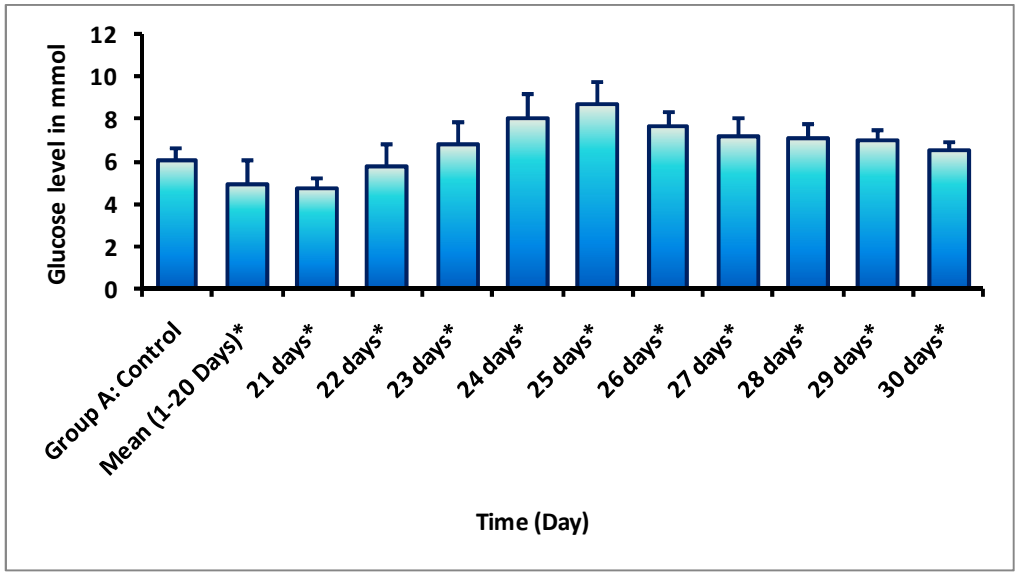

Fig. 4. Blood glucose level for prophylactic study after giving ayurvedic drug bohumutrantak ras in rats at dosage of 200mg/kg of body weight for 30 days followed by alloxan (150mg/kg body weight) from 21 to 30 days by intraperitoneal route. The standard deviations are shown as error bars. The differences in the curves are statistically significant (*p*=0.05) which have been shown by asterisks

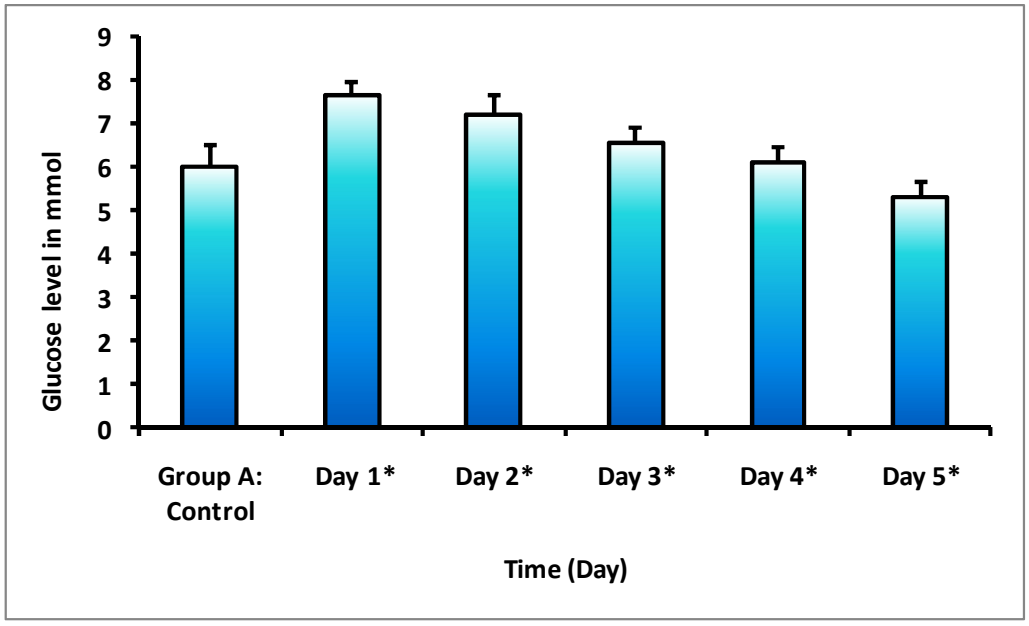

Fig. 5. Blood glucose level for prophylactic study after giving ayurvedic drug jambadyarista along with alloxan in rats for 5 consecutive days. The standard deviations are shown as error bars. The differences in the curves are statistically significant (*p*=0.05) which have been shown by asterisks

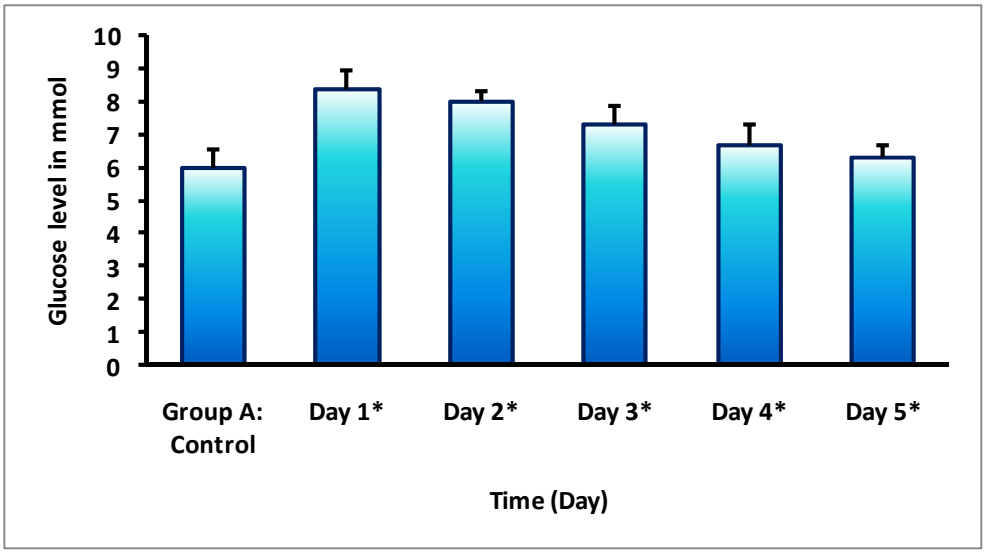

Fig. 6. Blood glucose level for prophylactic study after giving ayurvedic drug bohumutrantak ras along with alloxan in rats for 5 consecutive days. The standard deviations are shown as error bars. The differences in the curves are statistically significant (p=0.05) which have been shown by asterisks

4. DISCUSSION

Diabetes mellitus is the world's largest endocrine disease involving metabolic disorders of carbohydrate, fat and protein [12-14]. At present the treatment of diabetes mainly involves a sustained reduction in hyperglycemia by the use of biguanides, thiazolidinediones, sulfonylureas, *d*-phenylalanine and glycosidase-inhibitors in addition to insulin. However, due to unwanted side effects, the efficacies of these compounds are debatable and there is a demand for new compounds for the treatment of diabetes. Hence, plants have been suggested as a rich, yet unexplored source of antidiabetic drugs. The Ayurvedic medicines have a long history of treating diabetes. With a disturbing rise in the prevalence of the metabolic disease and associated healthcare cost, interest in complementary therapies has grown. Over the last 10-20 years, data from controlled investigations in animal models and patients have validated the therapeutic values of numerous Ayurvediotherapies for diabetes. The present study has been carried out to evaluate the prophylactic effects of the Jambadyarista and Bohumutrantak Ras on fasting blood glucose level on normal as well as alloxan-induced diabetic rats. Different dosages of Jambadyarista and Bohumutrantak Ras were used for proper investigation as standard antidiabetic effect in this study. As mentioned earlier, both Jambadyarista and Bohumutrantak Ras were administered consecutively for 20 days and from 21st day alloxan at a dose of 150 mg/kg and both Jambadyarista (in Group C) and Bohumutrantak Ras (in Group F) were administered simultaneously. Blood glucose levels were monitored. The values of blood glucose levels showed that alloxan failed to induce diabetes in Group C and Group F. We thus inferred that both Jambadyarista and Bohumutrantak Ras induced prophylaxis, the mechanism of which is still unknown, in rats of these groups. These results have showed that Jambadyarista and Bohumutrantak Ras significantly possess prophylactic effects against diabetes in alloxan-induced diabetic rats.

As a proposed mechanism it is assumed that alloxan as a cytotoxic agent causes a selective destruction of β-cell of islets of Langerhans and thus produced a marked decrease of insulin secretion from the β-cells**.** Jambadyarista and Bohumutrantak Ras contain different alkaloid that acts in the same manner and causes improvement in β-cells which might have the properties to stimulate or regenerate the β-cells for the secretion of insulin and are most effective for controlling diabetes.

Immunomodulation by herbal drugs is under being study by many research groups that may be useful in reducing the risk of various diseases and cancers. Although the mechanisms of some of the herbal drugs against the diseases are unclear and remain to be elucidated, they are important for further study to develop newly potential therapy agents for immunomodulation [15-19]. Recently, Ashwagandha (*Withania somnifera*, Family: *Solanaceae*) has been awarded for its use as vaccine adjuvant [20].

Thus, if it is possible to isolate the Ayurvedic active constituents from the reference drug, it will create a better opportunity for making new patented medicine for diabetic patients. The plant parts, therefore, seem to have promising value for the development of potent prophylactic Ayurvedic medicines for diabetes.

5. CONCLUSION

It can be inferred that the Ayurvedic drug “Jambadyarista and Bohumutrantak Ras” possess prophylactic activity against diabetes. They also showed antidiabetic effect in alloxan-induced diabetic rats. Further comprehensive cellular and molecular investigations are required to characterize the exact mechanism responsible for its prophylactic and antidiabetic effects.

CONSENT

Not applicable.

ETHICAL APPROVAL

All authors hereby declared that all experiments were examined and approved by the Ethical review committee, Faculty of Pharmacy, University Dhaka, Dhaka-1000, Bangladesh and were therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Manisha M, Priyanjali D, Jayant L, Saroj G, Thomas PAD. Indian herbs and herbal drugs used for the treatment of diabetes. J Clin Biochem Nutr. 2007;40(3):163-73.
2. Grover JK, Yadav S, Vats V. Medicinal plants of India with antidiabetic potential. J. Ethnopharmacol. 2002;81:81–100.
3. Griggs B. Green pharmacy: A history of herbal medicine. 1st ed. London: Robert Hale; 1981.
4. Evans JL. Diet, botanical, and nutritional treatments for type 2 diabetes. 2003. Accessed July 7, 2010.
5. Cusi K, DeFronzo RA. Metformin: A review of its metabolic effects. Diabetes Rev. 1998;6:89–31.
6. Marles RJ, Farnsworth NR. Antidiabetic plants and their active constituents. Phytomed. 1995;2:137–89.
7. Jung M, Park M, Lee HC, Kang YH, Kang ES, Kim SK. Antidiabetic agents from medicinal plants. Curr Med Chem. 2006;13:1203–18.
8. Prince PS, Menon VP, Pari L. Hypoglycemic activity of *Syzigium cumini* seeds: Effect on lipid peroxidation in alloxan diabetic rats. J Ethnopharmacol. 1998;61(1):1-7.
9. Available: www.herbaldestination.com/Natural_Health-Herbal_Supplement. Access on December 2013.
10. Sultana S, Haq M, Choudhuri, MSK, Sarker MH. In: Ghani, A. (ed.), Traditional Medicine, Jahangirnagar University, Dhaka, Bangladesh. 1990;90.
11. Chevallier A. The encyclopedia of medicinal plants, 1st edn., DK Publishing Inc., New York, USA. 1996;55-58;74;80;81;83;87;94-281.
12. Hossain MK, Sultan MZ, Rahman A, Uddin MG, Chowdhury AA, Rashid MA. RP-HPLC method development and validation for quantitative analysis of metformin and gliclazide in pharmaceutical formulations. J Sci Res. 2014;6(1):161-168.
13. Hossain K, Rahman A, Sultan MZ, Islam F, Akteruzzaman M, Salam MA, Rashid MA. A validated RP-HPLC method for simultaneous estimation of antidiabetic drugs pioglitazone HCl and glimepiride. Bangladesh Pharm J. 2013;16(1):69-75.
14. Akteruzzaman M, Rahman A, Sultan MZ, Islam F, Salam MA, Rashid MA. Development and validation of a simple RP-HPLC method for simultaneous estimation of metformin hydrochloride and rosiglitazone in pharmaceutical dosage forms. Dhaka Univ J Pharm Sci. 2012;11(2):157-163.
15. Huang CF, Lin SS, Liao PH, Young SC, Chi-Chiang Yang CC. The Immunopharmaceutical effects and mechanisms of herb medicine. Cell Mol Immu. 2008;5(1):23-31.
16. Tan BKH, Vanitha J. Immunomodulatory and antimicrobial effects of some traditional Chinese medicinal herbs: A review. Curr Med Chem. 2004;11;1423-1430.
17. Xiao-Yu L. Immunomodulating Chinese herbal medicines. Mem Inst Oswaldo Cruz Rio de Janeiro. 1991;86(Suppl.2):159-164.
18. Borchers AT, Hackman RM, Keen CL, Stern JS, Gershwin ME. Complementary medicine: A review of immunomodulatory effects of Chinese herbal medicines. Am J Clin Nutr. l997;66:1303-1312.

19. Tyler VE. Phytomedicines: Back to the future. J Nat Prod. 1999;62:1589-1592.
20. Composition of Ashwagandha and Indian Gooseberry, United States Patent. Patent. No.: US 2010/0098785 A1, Pub. Date: Apr. 22; 2010.

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