

Hypoglycemic Effect of Crude Methanolic Extract as Well as Sub Fractions of *Morus alba* on Rabbits

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Abstract: In the present research work the crude extract and its ethyl acetate and chloroform fractions of the leaves *Morus alba* were subjected for antidiabetic activity on selected rabbits having weight (1.2-2 kg). Alloxan monohydrate was used for induction of hyperglycemia. The samples to be tested were screened at 50, 100 and 200 mg/kg for their antidiabetic potential. The ethyl acetate fraction exhibited the most significant ($p < 0.01$) antidiabetic effect followed by crude methanolic extract and chloroform fraction. During 24 h study the significant effect was started from the 1st h of experiment which was remained upto the end of experiment (24h). The antidiabetic effect of our tested plant was comparable with the standard drug (metformin). The present research work strongly proved the antidiabetic effect of the leaves of *Morus alba*.

Key words: *Morus alba* • Alloxan monohydrates • Metformin and antidiabetic

INTRODUCTION

Diabetes mellitus is one of the metabolic disorders in other word we can say have close relation with abnormal metabolism and most commonly characterized by abnormal high blood glucose level due to inability of the body to properly utilize blood glucose level this is because of some defect at level of insulin [1]. Commonly diabetes mellitus have two types that is IDDM, insulin dependent diabetes mellitus and NIDDM that is non insulin dependent diabetes mellitus [2]. In case of NIDDM the beta cell of pancreases function become impaired due to insulin resistance leading to abnormal high blood glucose level [3, 4]. Diabetes mellitus is considered as one of the major risk factor for cardiovascular diseases such as stroke, heart attack, atherosclerosis etc [5]. Various drugs like insulin and oral hypoglycemic agents are used for treatment of diabetes mellitus any how these drugs have a lot number of side effect like, weight gain, hypoglycemic shock, lactic acidosis and almost all these drugs can cause liver and renal problems. Before development of new pharmaceutical products physician were totally

dependent on various herbal plants and also in present time they are in search of such products have good safety and efficacy profile [6]. It is cleared from a well known report that more than 800 medicinal plants are used as traditional remedies for treatment of diabetes throughout the world⁷. The plant species that is *Morus alba* widely distributed throughout the world belongs to a well known family that is *sterculiaceae*. Traditionally *Morus alba* is used in condition like, diabetes, convulsions related to epilepsy, mental problems, asthma, rheumatism and as anti-inflammatory [8-10].

The main objective of present study is to observed the effect of crude extract as well as subtractions of the leaves of *Morus alba* on selected rabbits where diabetes was induced with help of alloxan.

MATERIALS AND METHODS

Extract Preparation: The plant leaves were collected, Shad dried and pulverized and dried powder plant materials were obtained. After powder maceration, crude methanolic extract was obtained according to well establish reported protocols [11-17]. After filtration and

concentration under vacuum at 40°C, 300 g crude methanolic extract was obtained. The extract was further fractioned with various solvents on the basis of polarity such as (*n*-hexane, chloroform, ethyl acetate, *n*-butanol and aqueous fractions). Selected number of rabbits were categorized in various groups according to well establish procedure under control laboratory condition accordingly.

Experimental Procedure: In experimental procedure diabetes in the respective rabbits were produced with help of alloxan and rabbits were fasted for 18 hrs before experimental process and animals were divided in various groups and each group contain respective number of rabbits according to procedure. The first group served as a control which only received saline the second group served as a diabetic control and received alloxan monohydrate the third group received metformin as a standard drug where other groups rabbits were treated with various crude extract and subtraction in a specific doses that is 50,100 and 200 respectively .Diabetes was produced with help of alloxan administered intravenously [18]. Once 48 hrs elapsed blood was collected to check for blood glucose level and those animals were selected have blood glucose level more than 200mg/dl and were expose to various extract and fractions and blood glucose level was properly monitored with help of glucometer with intervals of 0.5 hrs, 1 hrs, 2 hrs, 4 hrs, 6 hrs, 8 hrs,10 hrs,12 hrs and 24 hrs.

Antidiabetic Activity: Antidiabetic activity was performed on selected animals having blood glucose level more than 200 mg /dl animals were kept in fasting condition for 18

hours and on the day of experiment the respective doses of crude extract, ethyl acetate and chloroform were applied to the respective animals and after specific period of time blood was withdrawn with specific intervals to check for blood glucose levels with help of glucometer as shown in Tables.

Statistical Analysis: For statistical analysis, ANOVA was followed by post hoc Dunnetts test for multiple comparisons. Effects were considered to be significant at the $P < 0.05$ level.

RESULTS

The crude methanolic extract of the leaves of our selected plant demonstrated a significant antidiabetic effect at the tested dose of 50, 100 and 200 mg/kg. the effect was dose dependant as well time dependent, because the crude methanolic extract significantly reduced the induced hyperglycemia after 1st h and this effect was increased with the passage of time as presented in Table 1. The percent reduction of hyperglycemia by methanolic extract (50 mg/kg) was 7.8, 10.18, 13.39, 11.57, 9.15, 6.08, 5.26 and 4.30 after 0.5, 1, 2, 4, 6, 8, 10, 12 and 24 respectively. This percent reduction of hyperglycemia was approved with increasing the dose. In case of ethyl acetate fraction, a dose dependant effect was observed as give in Table 2. The percent reduction of glucose level at 200 mg/kg was 13.5, 15.25, 17.54, 18.30, 20.57, 22.84, 22.47 and 26.08 after 0.5, 1, 2, 4, 6, 8, 10, 12 and 24 respectively. The effect of chloroform fraction was similar to crude methanolic extract as clear from Table 3.

Table 1: Percent blood glucose reduction by *Morus alba* methanolic

Groups	Dose/kg	IBG	Percent blood glucose reduction (mg/dl) in hours								
			0.5	1	2	4	6	8	10	12	24
Control	10 ml	224.7	0.8	0.48	0.06	0.34	0.34	0.13	0.41	0.48	0.13
Methanolic	50 mg	216	7.8*	10.18*	13.39*	11.57*	9.15*	6.08*	5.26*	4.30*	3.00
	100 mg	204.9	9.7*	11.25*	14.91*	16.41**	18.68**	18.41**	21.10**	17.33**	13.03**
	200 mg	219.2	11.5**	14.35**	16.56**	18.26**	19.57**	20.87**	22.45**	25.06***	21.83***
Metformin	0.5 mg	223.9	15.6 ***	19.74***	24.12***	19.83**	17.14**	17.1**	14.72**	12.23**	8.60**

IBG: Initial Blood Glucose (mg/dl), * $P < 0.05$, ** $P < 0.01$, ***. $P < 0.001$

Table 2: Percent blood glucose reduction by *Morus alba* ethyl acetate fraction

Groups	Dose/kg	IBG	Percent blood glucose reduction (mg/dl) in hours								
			0.5	1	2	4	6	8	10	12	24
Control	10 ml	224.7	0.8	0.48	0.06	0.34	0.34	0.13	0.41	0.48	0.13
Ethyl acetate	50 mg	216	3.3	8.2*	11.15*	14.35*	12.57*	10.20*	7.08*	5.28*	6.30*
	100 mg	230.2	10.5**	12.27**	16.71**	17.46**	19.60**	20.40**	22.18**	18.35**	14.03**
	200 mg	245.3	13.5**	15.25**	17.54**	18.30**	20.57***	22.84***	22.47***	26.08***	23.83***
Metformin	0.5 mg	223.9	15.6 ***	19.74***	24.12***	19.83**	17.14**	17.1**	14.72**	12.23**	8.60**

IBG: Initial Blood Glucose (mg/dl), * $P < 0.05$, ** $P < 0.01$, ***. $P < 0.001$

Table 3: Percent blood glucose reduction by *Morus alba* chloroform fraction.

Groups	Dose/kg	IBG	Percent blood glucose reduction (mg/dl) in hours								
			0.5	1	2	4	6	8	10	12	24
Control	10 ml	224.7	0.8	0.48	0.06	0.34	0.34	0.13	0.41	0.48	0.13
Chloroform.	50 mg	216	7.12*	10.17*	13.40*	11.57*	8.15*	7.08*	4.26	3.30	3.00
	100 mg	204.9	10.22*	12.11**	15.55**	17.20**	18.80**	18.24**	21.7**	16.33**	13.03*
	200 mg	219.2	12.12**	15.32**	16.86**	18.89**	20.57***	21.67***	22.25***	25.06***	21.83***
Metformin	0.5 mg	223.9	15.6***	19.74***	24.12***	19.83**	17.14**	17.1**	14.72**	12.23**	8.60**

IBG: Initial Blood Glucose (mg/dl), * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

DISCUSSION

As we know that beta cell of pancreas in our body responsible for normal secretion of insulin that causing proper utilization of blood glucose level responsible for normal body functions. Any mutation or disturbances at level of beta cell of pancreas can cause abnormal or even in some condition ceased the secretion of insulin and the effect is observed in the form of IDDM or NIDDM which if not properly investigate or diagnosis have a lot of complications and even death.

Alloxan monohydrate has a direct effect on beta cell of pancreases and causing its destruction by causing necrosis of beta cells of pancreases this cytotoxic action of alloxan on beta cell is due to reactive oxygen species have direct relation to alloxan. Experimental it is cleared that *Morus alba* leaves crude methanolic extract and fractions showed significant anti diabetic activity in other word have direct effect on decreasing blood glucose level in mentioned dose.

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