ACUTE TOXICITY AND HYPOGLYCAEMIC ACTIVITY OF 70% ETHANOLIC EXTRACT OF *MIRABILIS JALAPA* L.

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Abstract

Mirabilis jalapa L. is locally known as Mye-su, Lay-nar-yi-pan, Marvel of Peru or Four o'clock flower and belongs to the family Nyctaginaceae. The plant samples were collected from Kamaryut Township, Yangon University Campus. In acute toxicity study of 70% ethanolic extract of *Mirabilis jalapa* L. were evaluated on albino mice by using method of OECD Guideline 423. There were no sign of toxicity and lethality of mice, even with the maximum dose at 5 g/kg body weight of the extract during the test period of 14 days. Therefore, 70% ethanolic extract of *Mirabilis jalapa* L. had no acute toxic effect up to the dose of 5 g/kg. The hypoglycaemic activity of 70% ethanolic extract was also studied on adrenaline-induced hyperglycaemic rats model by using the method of Gupta *et al*. (1967). In hypoglycaemic activity test on adrenaline-induced hyperglycaemic rats, the extract at the dose of 1 g/kg showed significant blood glucose lowering effect at 4 hrs ($p<0.01$), 2 g/kg at 3 hrs ($p<0.005$) and 4 hrs ($p<0.001$) and 4 g/kg at 2 hrs, 3hrs and 4 hrs ($p<0.001$) when compared with the control. It was found that 4 g/kg of 70% ethanolic extract showed the most effective hypoglycaemic activity among 3 doses of the extract.

Keywords: *Mirabilis jalapa* L., Acute Toxicity, Hypoglycaemic Activity

Introduction

Mirabilis jalapa L. belongs to the family Nyctaginaceae. It consists of about 30 genera and 300 species in tropical and subtropical regions of both the Old and New World (Cronquist, 1981 and Bhattacharyya, 1998). The native of this plant is tropical America (Cooke, 1958).The world is now moving towards the herbal medicine or phytomedicines that repair and strengthening bodily systems and help to destroy offending pathogens without toxic side effects. Today, it has been developed as a separate industry as many people favor herbal medicine over synthetic medicine (Pandey *et al*., 2011).

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Treatment of diseases like cancer, diabetes etc. is not easy for the poor family due to high coast of the treatment. Nowadays, there is widespread interest to promote the traditional health care systems to meet primary health care needs (Pallab *et al*., 2014).

Toxicity is an expression of being poisonous, indicating the state of adverse effects led by the interaction between toxicants and cells. This interaction may vary depending on the chemical properties of the toxicants and the cell membrane, as it may occur on the cell surface, within the cell body, or in the tissues beneath as well as at the extracellular matrix. The toxic effects may take place prior to the binding of the toxicants to the vital organs such as liver and kidneys. Hence, evaluation of toxic properties of a substance is crucial when considering for public health protection because exposure to chemicals can be hazardous and results to adverse effects on human being. (Chen *et al*., 2011).The acute toxicity test in which single dose of the drug is used in each animal on one occasion only for the determination of gross behavior and LD_{50} (the dose which has proved to be lethal causing death to 50% of the tested group of animals) or median lethal dose (Gupta, 2012).

There is hundreds of other bioactive compounds present in plants are helpful for the treatment of diabetic diseases and also used in the lowering of glucose level in the blood (Marles and Farnsworth, 1995). Traditionally plants are also used for the treatment of diabetes throughout the world. Management of diabetes without any side effect is still a challenge for the medical system. This leads to an increasing search for improved antidiabetic drugs (Sarkar *et al*., 2011).

Diabetes is a serious, chronic disease that occurs either when the pancreas does not produce enough insulin (a hormone that regulates blood glucose), or when the body cannot effectively use the insulin it produces. Raised blood glucose, a common effect of uncontrolled diabetes, may, over time, lead to serious damage to the heart, blood vessels, eyes, kidneys and nerves. More than 400 million people live with diabetes. There are two types of diabetes.

Type 1 diabetes is characterized by deficient insulin production in the body. People with type 1 diabetes require daily administration of insulin to regulate the amount of glucose in their blood. If they do not have access to insulin, they cannot survive. The majority of type 1 diabetes occurs in children and adolescents. Symptoms include excessive urination and thirst constant hunger, weight loss, vision changes and fatigue.

Type 2 diabetes results from the body's ineffective use of insulin. Several dietary practices are linked to type 2 diabetes risk, including high intake of saturated fatty acids, high total fat intake and inadequate consumption of dietary fibre, sugar-sweetened beverages, overweight and obesity, unhealthy diet, physical inactivity and also active (as distinct from passive) smoking to increase risk. Diabetes can damage the heart, blood vessels, eyes, kidneys and nerves, leading to disability and premature death (WHO, 2016).

Nowadays, several antidiabetic agents are avaliable but most have certain adverse effects and high cost. The majority of herbal medicines seem to have efficacy, low incidence of serious adverse effects and low cost. The use of herbal medicine is increasing in both developing and developed countries due to growing recognition of natural products and easy availability at affordable prices and sometimes the only source of health care available for rural people (Atmakuri, 2010).

The aim of the present research is to find the medicinal value of *Mirabilis jalapa* L. plant and to promote the intensive application of Myanmar traditional medicine. Screening the acute toxicity and hypoglycaemic activity of 70% ethanolic extracts from *Mirabilis jalapa* L. plant were investigated.

Materials and Methods

Acute toxicity test of 70% ethanolic extract of *Mirabilis jalapa* L. plant on albino mice

The acute toxicity test on mice was carried out according to the method of OECD Guideline 423 (2001). Adult, 18 albino mice, Dutch Denken Yoken strain of female sex, weighting between 25- 30 g, were used for acute

toxicity study. These animals were provided by the Laboratory Animal Services Division, Department of Medical Research, and Yangon.

Materials

Test animals	-	18 female albino mice (ddy strain, body weight 25- 30g)
Test agents	-	Distilled water, 70% ethanol extract from *Mirabilis jalapa* L. plant
Apparatus	-	Mice cages, animal balance, "18" gague intragastric needle, disposal syrings (1ml and 5 ml), rubber gloves and masks
Dose	-	2g/kg and 5g/kg (body weight) of albino mice
Period of observation	-	14 days

Methods

Acute toxicity Test of 70% ethanolic extracts of *Mirabilis jalapa* L. plant was evaluated by the methods of OECD Guidelines 423. The female albino mice were randomly selected and kept in their cages for at least 5 days prior to dosing to allow for acclimatization to the laboratory conditions. According to the test description, total number of 18 female albino mice (ddy strain), weighing between 25- 30 g were selected and divided into three groups. Each group contained six mice and kept in the each mouse cages. At first, the mice were individually marked on the parts of the body and weighed. Required doses based on the body weight of the mice were calculated. The mice were fasted for 18 hours before experiment but were allowed with free access to water. Group (I) mice served as a control group and they were administered 10 ml/kg of distilled water orally. In this study, starting dose 2 g/kg was chosen. The extract was dissolved in distilled water and the required doses were administered orally by using intragastric needle to every mice. A limited test at one dose level of 2 g/kg was carried out with 6 mice (3 mice per step). After administration of the test agent orally, they were allowed to have

food and water. The sign of toxicity such as changes in skin and fur, eyes, salivation, convulsion, cyanosis, tremors and diarrhoea or lethality were observed on test animals. Treated mice were observed individually after dosing at one time during the first 30 minutes hourly up to 4 hours for first 24 hours. After that, all mice were monitored daily up to 14 days.

Thus, another 6 mice (3 mice per step) were administered 5 g/kg. The mice were observed for toxic sign by using the method described above. All the mice were observed to detect the delayed toxicity up to 14 days. The mortality and toxic signs during this period were noted.

Acute toxicity test of 70% ethanolic extract of *Mirabilis jalapa* L. plant on albino mice

Figure 1. Mice cages (each contains 6 mice)

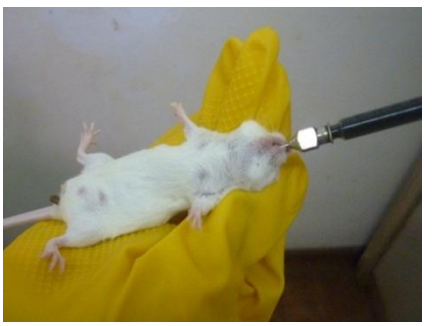

Figure 2. Administration of extract suspension to mice

Hypoglycaemic activity of 70% ethanolic extract of *Mirabilis jalapa* L. plant on adrenaline-induced hyperglycaemic rat model

The hypoglycaemic activity of 70% ethanolic extract was also studied on adrenaline-induced hyperglycaemic rats model by using the method of Gupta *et al*. (1967) at Department of Medical Research (DMR), Yangon.

Materials

Test animals - 8 Wistar strain albino rats of both sexes (body weight 180- 250 g)

Test agent	-	Distilled water, 70% ethanolic extract, Glibenclamide tablets 5 mg (Malaysia), Adrenaline injection (1 mg/ml) (Myanmar Pharmaceutical Factory)
Apparatus	-	Aluminium cages, Animal balance, Spirit cotton wools, disposable syringes with needle (1 ml, 5 ml), Glucometer, Test strips, 18 gauge dosing needle, rubber gloves and masks
Dose Schedule	-	70% ethanolic extract (1 g/kg, 2 g/kg and 4 g/kg) body weight

Methods

Test animal profile

The study of hypoglycaemic effect of 70% ethanolic extract of *Mirabilis jalapa* L. plant was performed by using the method of Gupta *et al*. (1967). Both sexes of eight adult healthy albino rats of Wistar strains weighing (180- 250 g) obtained from Department of Medical Research were used in this experiment. They were kept in clean and dry cages to allow for acclimatization to the laboratory conditions one week before starting the experiment. The rats were fasted overnight for 18 hours before the experiment but water was allowed freely. Firstly, they were served as control group and only distilled water was given orally to them during experiment.

Preparation and administration of drug suspension

Before the experiment, individual rats were marked, weighed and kept without food for 18 hours. The dosage was calculated according to the body weight of rat. Control animals were administered orally with 10 ml/kg of distilled water. The drug suspension (i.e. distilled water) was given orally to each rat by using an intragastric needle connected to a plastic syringe containing the calculated dosage. The syringe was put into the stomach. Then, the piston was pushed to deliver the test agents into the stomach. Immediate sneezing and coughing indicated injecting into the lungs and in such condition, the syringe was withdrawn.

Collection of blood sample and induction of hyperglycaemia in rats

Before the drug administration, the blood sample was collected by cutting about 1 mm at the tip of the tail as the base line blood sample (0hr). The glucometer test strip was inserted into the glucometer and then, one drop of the blood sample was dropped on this strip. Blood glucose concentration was measured by glucometer at 0 hour. The results were expressed in (mg/dl). Then, these rats were orally given with distilled water (10 m/kg) by using "18" gauge intragastric needle. Thirty minutes after administration of distilled water, these rats were subcutaneously injected with (0.4 ml/kg) body weight of adrenaline to the back of the neck. Then, bloods were taken from tail vein and blood glucose levels were determined hourly up to 4 hours with glucometer. After taking the blood sample, the tail of the rat was rubbed with cotton wool soaked in absolute alcohol to protect the puncture against infection.

Determination of hypoglycaemic activity of 70% ethanolic extract (1 g/kg, 2 g/kg and 4 g/kg body weight) of *Mirabilis jalapa* L. plant on adrenaline-induced hyperglycaemic rats

After one week washout period, the same 8 rats were used again and these rats were kept without food for 18 hours before experiment. Only water was allowed orally to them. After that, these rats were orally given ethanolic extract (1 g/kg) body weight by using "18" gauge intragastric needle. After 30 minutes, these rats were subcutaneously injected with (0.4 ml/kg) body weight of adrenaline. Fasting blood was taken from tail vein and blood glucose levels were determined at 0hr, 1hr, 2hr, 3hr and 4 hours with glucometer. Then, all the animals were allowed to rest for one week of drug free period (i.e. washout period). After washout period for one week, the same 8 rats were also tested for determination of blood glucose level with 70% ethanolic extract, (2 g/kg) body weight. Determinations of blood glucose levels were performed as above procedures.

After washout period of one week, the same 8 rats were tested with 70% ethanolic extract, (4 g/kg) body weight for determination of blood glucose level as above procedures.

Determination of hypoglycaemic activity of standard drug, (glibenclamide) on adrenaline-induced hyperglycaemic rats

After drug free interval of one week, the same 8 rats were used again and these rats were kept without food for 18 hours before experiment. Fasting blood glucose levels (0hr) were taken from venous blood obtained by cutting about 1 mm at the tip of the tail and measured by glucometer. After that, these rats were orally given with standard drug glibenclamide (4 mg/kg) body weight by using "18" guage intragastric needle. After 30 minutes, these rats were subcutaneously injected with (0.4 ml/kg) body weight of adrenaline. Then, bloods were taken from tail vein hourly at 1hr, 2hr, 3hr and up to 4 hours and determination of blood glucose levels were done with glucometer.

The study design used in this study was cross over study design in albino rats.

Data management and analysis

Standard statistical methods were used in the calculation of arithmetic mean (X) standard deviation (SD) and standard error (SE). Paired student "t" test were used to analyze the significant differences between means of control and experimental groups (Gupta *et al*., 1967).

Determination of blood glucose concentration

Percent reduction was calculated by the following formula;

$$\text{Percent reduction} = \frac{\text{Difference between rises in blood glucose level of control and test}}{\text{Blood glucose level rise in control}} \times 100$$

$$= \frac{C-T}{C} \times 100$$

C = rise in blood glucose level of control

T = rise in blood glucose level of test

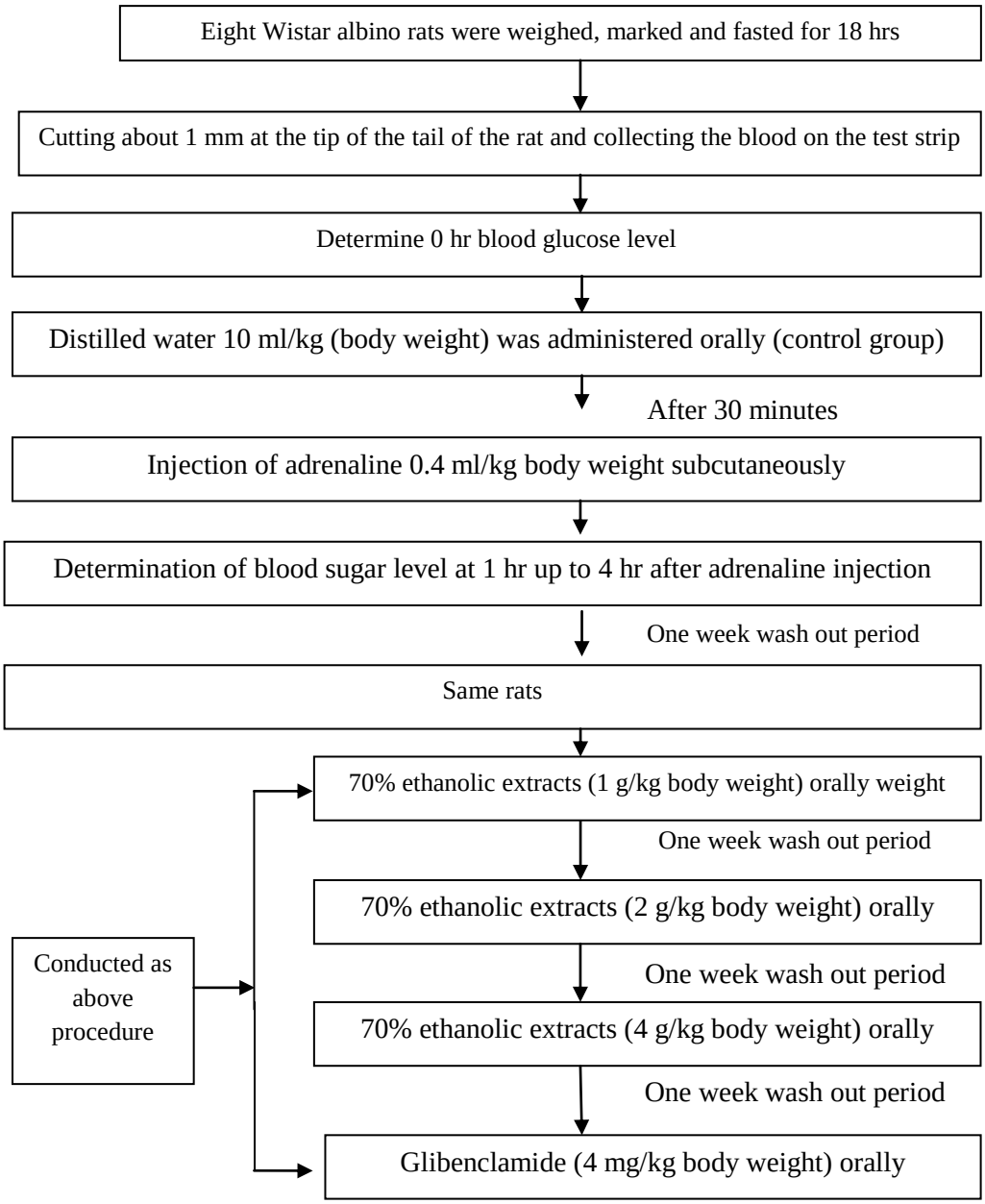

Figure 3. Flow chart for hypoglycaemic activity testing in adrenaline-induced hyperglycaemic rats

Hypoglycaemic activity of 70% ethanolic extract of *Mirabilis jalapa* L. plant on adrenaline-induced hyperglycaemic rat model

Figure 4. Albino rats in cages and each contains 2 rats

Figure 5. Cutting the tip of tail from the rats

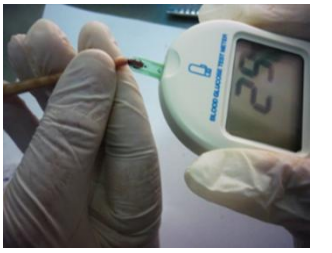

Figure 6. Determination of blood glucose level by using glucometer

Figure 7. Administration of distilled water to rat

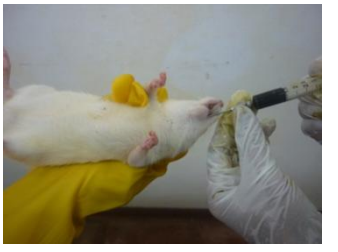

Figure 8. Administration of extracts suspension to rat

Figure 9. Adrenaline injection into nape of neck of albino rat

Results

Acute toxicity test of 70% ethanolic extract of *Mirabilis jalapa* L. plant on albino mice

In this study, the mice were administered with the dose of 2 g/kg (body weight) and 5 g/kg (body weight) of 70% ethanolic extract of *Mirabilis jalapa* L. Each group of mice was still alive and did not show any signs of toxicity in skin, fur and eyes. Salivation, convulsion, cyanosis, tremors and diarrhoea were not detected. Even with the maximum dose of 5 g/kg body weight of 70% ethanolic extracts administration, there was no lethality and toxic effect

up to 14 days of observation period. Therefore, it was observed that median lethal dose (LD_{50}) of the extract was more than 5 g/kg and the extract was not toxic up to the dose of 5 g/kg. The results were shown in Table (1).

Table 1. Acute toxicity test of 70% ethanolic extract of *Mirabilis jalapa* L. plant on albino mice

No. of Group	Type of drug administration	No. of mice tested	Dosage	Observed period	No. of death
I	Control (distilled water)	6	10 ml/kg	14 days	0/6
II	70% ethanolic extracts	6	2 g/kg	14 days	0/6
III	70% ethanolic extracts	6	5 g/kg	14 days	0/6

The mice were found to be alive and healthy during two weeks. No lethality and toxic effect of the mice were observed up to 14 days. These extracts were free from acute toxic effect at the doses 2 g/kg and 5 g/kg.

Hypoglycaemic activity of 70% ethanolic extract of *Mirabilis jalapa* L. plant on adrenaline-induced hyperglycaemic rat model

The hypoglycaemic activity of 70% ethanolic extracts of *Mirabilis jalapa* L. was tested by using adrenaline induced hyperglycaemic albino rats. Eight adult healthy Wistar strain albino rats of both sexes, weighing between (180-250 g) body weight were used for this study. The results of hypoglycaemic activity were shown in Tables (2 - 3) and Figures (10 -11).

Effect of distilled water on blood glucose levels on adrenaline-induced hyperglycaemic rats model (control group)

The mean blood glucose level of the 8 albino rats given orally with distilled water (10 ml/kg body weight) at 0 hr, 1 hr, 2 hr, 3 hr and 4 hr after subcutaneous injection of adrenaline 0.4 ml/kg were 62.00 ± 2.62 mg/dl, 191.13 ± 8.93 mg/dl, 249.25 ± 9.77 mg/dl, 226.88 ± 8.52 mg/dl and 210.38 ± 7.4 mg/dl respectively. It was found that blood glucose level significantly increased at 1 hr, 2 hr, 3 hr and 4 hr after injection of adrenaline (0.4 ml/kg) as shown in Table (2).

Effect of different doses levels of 70% ethanolic extract (1 g/kg, 2 g/kg and 4 g/kg body weight) of *Mirabilis jalapa* L. plant on blood glucose level on adrenaline-induced hyperglycaemic rats model

The mean blood glucose level of the 8 albino rats treated with 70% ethanolic extracts of *Mirabilis jalapa* L. **(**1 g/kg body weight) at 0 hr, 1 hr, 2 hr, 3 hr and 4 hr after subcutaneous injection of adrenaline (0.4 ml/kg) were 77.00 ± 3.96 mg/dl, 199.63 ± 5.55 mg/dl, 250.63 ± 6.11 mg/dl, 252.75 ± 10.69 mg/dl and 222.38 ± 11.75 mg/dl respectively. It was observed that the oral administration at 70% ethanolic extracts of *Mirabilis jalapa* L. (1 g/kg body weight) produced a significant decrease in glucose level at 4 hr ($p<0.01$) when compared with that of control group as shown in Table (2) and Figure (10).

The mean blood glucose level of the 8 albino rats treated with 70% ethanolic extracts of *Mirabilis jalapa* L. **(**2 g/kg body weight) at 0 hr, 1 hr, 2 hr, 3 hr and 4 hr after subcutaneous injection of adrenaline (0.4 ml/kg) were 74.63 ± 3.48 mg/dl, 205.00 ± 4.47 mg/dl, 253.25 ± 9.11 mg/dl, 227.75 ± 10.17 mg/dl and 201.00 ± 8.91 mg/dl respectively. It was observed that the oral administration of 70% ethanolic extracts of *Mirabilis jalapa* L. (2 g/kg body weight) produced a significant decrease in glucose level at 3 hr ($p<0.005$) and 4 hr ($p<0.001$) when compared with that of control group as shown in Table (2) and Figure (10).

The mean blood glucose level of the 8 albino rats treated with 70% ethanolic extracts of *Mirabilis jalapa* L. **(**4 g/kg body weight) at 0 hr, 1 hr, 2 hr, 3 hr and 4 hr after subcutaneous injection of adrenaline (0.4 ml/kg) were 63.75 ± 2.17 mg/dl, 201.63 ± 5.84 mg/dl, 223.50 ± 5.61 mg/dl, 199.13 ± 5.7 mg/dl and 178.25 ± 5.51 mg/dl respectively. It was observed that the oral administration at 70% ethanolic extracts of *Mirabilis jalapa* L. (4 g/kg body weight) produced a significant decrease in glucose level at 2 hr ($p<0.001$), 3 hr ($p<0.001$) and 4 hr ($p<0.001$) when compared with that of control group as shown in Table (2) and Figure (10).

Effect of standard drug, glibenclamide on blood glucose level in adrenaline-induced hyperglycaemic rats model

The results of mean blood glucose level of the 8 albino rats treated with standard drug glibenclamide (4 mg/kg body weight) at 0 hr, 1 hr, 2 hr, 3 hr and 4 hr after subcutaneous injection of adrenaline 0.4 ml/kg were 72.00 ± 2.34 mg/dl, 149.25 ± 7.93 mg/dl, 193.13 ± 6.58 mg/dl, 172.63 ± 15.38 mg/dl and 114.38 ± 4.24 mg/dl respectively. The results of the oral administration of standard drug, glibenclamide showed that the blood glucose level of adrenaline-induced rats were significant decreased at 1 hr ($p<0.05$), 2 hr ($p<0.001$), 3 hr ($p<0.001$) and 4 hr ($p<0.001$) when compared with that of control group are shown in Table (2) and Figure (10).

Comparison of percent reductions of blood glucose level with different dose of 70% ethanolic extract from *Mirabilis jalapa* L. plant and standard drug, glibenclamide.

The comparison of mean percent reductions of blood glucose levels with 70% ethanolic extracts from *Mirabilis jalapa* L. and standard drug, glibenclamide are shown in Table (3) and Figures (11).

The mean percent reduction of blood glucose level with 70% ethanolic extracts (1 g/kg body weight) were 2.23 ± 6.47%, 6.35 ± 1.75%, 8.95 ± 2.88% and 14.94 ± 3.57% at 1 hr, 2 hr, 3 hr and 4 hr respectively. The mean percent reduction of blood glucose level with 70% ethanolic extracts (2 g/kg body weight) were -5.39 ± 10.15%, 3.92 ± 4.03%, 21.24 ± 4.23% and 26.18 ± 4.48% at 1 hr, 2 hr, 3 hr and 4 hr respectively. The mean percent reduction of 70% ethanolic extracts (4 g/kg body weight) were -10.43 ± 8.63%, 13.45 ± 2.2%, 28.73 ± 3.94% and 31.64 ± 3.11% at 1 hr, 2 hr, 3 hr and 4 hr respectively. The mean percent reduction of blood glucose level with glibenclamide (4 mg/kg body weight) were 33.91 ± 12.15%, 33.99 ± 4.19%, 47.89 ± 6.61% and 60.59 ± 6.72% at 1 hr, 2 hr, 3 hr and 4 hr respectively.

Table 2. Mean blood glucose concentration (Mean ± SE) of 70% ethanolic extract of *Mirabilis jalapa* L. plant (1 g/kg, 2 g/kg, 4 g/kg) and glibenclamide, (4 mg/kg) on adrenaline- induced hyperglycaemic rats model

Group of rats	Blood glucose concentration (mg/dl)				
	0 HR	1 HR	2 HR	3 HR	4 Hr
Control	62.00 ± 2.62	191.13 ± 8.93	249.25 ± 9.77	226.88 ± 8.52	210.38 ± 7.4
70% ethanolic extract 1 g/kg	77.00 ± 3.96	199.63 ± 5.55	250.63 ± 6.11	252.75 ± 10.69	222.38 ± ** 11.75
70% ethanolic extract 2 g/kg	74.63 ± 3.48	205.00 ± 4.47	253.25 ± 9.11	227.75 ± *** 10.17	201.00 ± **** 8.91
70% ethanolic extract 4 g/kg	63.75 ± 2.17	201.63 ± 5.84	223.50 ± **** 5.61	199.13 ± **** 5.7	178.25 ± **** 5.51
Glibenclamide 4 mg/kg	72.00 ± 2.34	149.25 ± * 7.93	193.13 ± **** 6.58	172.63 ± **** 15.38	114.38 ± **** 4.24

*P < 0.05, **P < 0.01, ***P < 0.005, ****P < 0.001

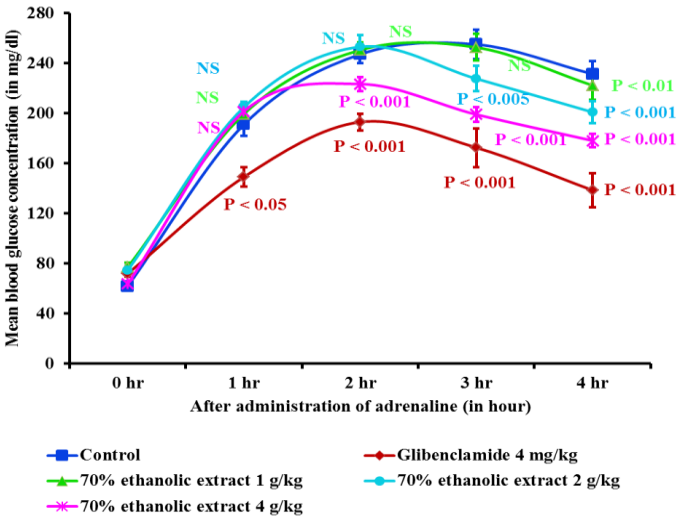

Figure 10. Time course of the effect of 70% ethanolic extracts of *Mirabilis jalapa* L. plant (1 g/kg, 2 g/kg and 4 g/kg) and glibenclamide, (4 mg/kg) on adrenaline- induced hyperglycaemic rats model

Table 3. Percent reduction (Mean ± SE) of hyperglycaemic with 70% ethanolic extract of *Mirabilis jalapa* L. plant and glibenclamide (4 mg/kg) on adrenaline-induced hyperglycaemic rats model

Group of rats	Percent reduction of hyperglycaemic			
	1 HR	2 HR	3 HR	4 HR
Glibenclamide 4 mg/kg	33.91 ± 12.15	33.99 ± 4.19	47.89 ± 6.61	60.59 ± 6.72
70% ethanolic extract 1 g/kg	2.23 ± 6.47	6.35 ± 1.75	8.95 ± 2.88	14.94 ± 3.57
70% ethanolic extract 2 g/kg	-5.39 ± 10.15	3.92 ± 4.03	21.24 ± 4.23	26.18 ± 4.48
70% ethanolic extract 4 g/kg	-10.43 ± 8.63	13.45 ± 2.2	28.73 ± 3.94	31.64 ± 3.11

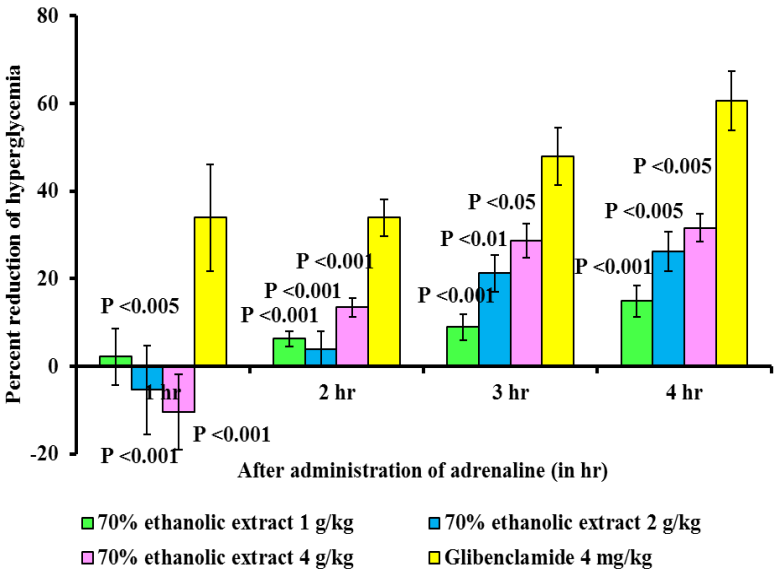

Figure 11. Percent reduction of hyperglycaemic with 70% ethanolic extract of *Mirabilis jalapa* L. plant (1 g/kg, 2 g/kg and 4 g/kg) and glibenclamide, (4 mg/kg) on adrenaline- induced hyperglycaemic rats model. N=8, each point represents the mean of observations and the vertical bars indicate standard errors of the means.

Discussion and Conclusion

In this study, acute toxicity test of 70% ethanolic extract of *Mirabilis jalapa* L. plant was evaluated by using the methods of OECD Guidelines 423. The mice were administered with the dose of 2 g/kg (body weight) and 5 g/kg (body weight) of 70% ethanolic extract of *Mirabilis jalapa* L. All the animals did not show any signs of toxicity and lethality in observation period of 14 days. There were no toxic signs and leathality during the observation period of 14 days with 2 g/kg (body weight) and the maximum dose of 5 g/kg (body weight). Therefore, 70% ethanolic extract of *Mirabilis jalapa* L. plant had no acute toxic effect up to the dose of 5 g/kg.

Prakash *et al*. (2012) reported that the hydro-alcoholic extract of aerial parts of *Mirabilis jalapa* L. possessed antihyperglycaemic activity at dose levels of 200 mg/kg and 400 mg/kg in streptozotocin induced hyperglycaemic animals.

Doss *et al*. (2015) reported that the hydro-ethanolic leaf extract of *Mirabilis jalapa* L. possessed potent antidiabetic activity in streptozotocin induced diabetic rats.

In this study, the hypoglycaemic effect of 70% ethanolic extract of *Mirabilis jalapa* L. plant at the dose of (1 g/kg, 2 g/kg and 4 g/kg) were investigated on adrenaline induced hyperglycaemic rats model by using the method of Gupta *et al*. (1967). 70% ethanolic extract (1 g/kg) significantly decreased the blood glucose concentration of the rats at 4 hour ($p<0.01$) after subcutaneous injection of adrenaline. 70% ethanolic extract (2 g/kg) significantly decreased the blood glucose concentration of the rats at 3 hour ($p<0.005$) and 4 hour ($p<0.001$) after subcutaneous injection of adrenaline. 70% ethanolic extract (4 g/kg) significantly decreased the blood glucose level at 2 hour up to 4 hour ($p<0.001$) after subcutaneous injection of adrenaline. It was found that 4 g/kg of 70% ethanolic extract was more effective than 1 g/kg and 2 g/kg doses. Therefore, hypoglycaemic effects of 70% ethanolic extract from *Mirabilis jalapa* L. plant was found to be in dose dependent manner.

In this study, the hypoglycaemic effects of standard drug, glibenclamide at the dose level of 4 mg/kg showed a significant reduction in

blood glucose level at 1 hour ($p<0.05$) and at 2 hour, 3 hour and 4 hour ($p<0.001$) after the administration of drugs on adrenaline induced hyperglycaemic rats. In this study, 70% ethanolic extract of *Mirabilis jalapa* L. plant at the doses of 1 g/kg, 2 g/kg and 4 g/kg showed significant reduction of blood glucose level on adrenaline induced hyperglycaemic rats.

At 1 hour after subcutaneous injection of adrenaline, mean percent reductions of hyperglycaemia with 70% ethanolic extract of *Mirabilis jalapa* L. plant at the dose of 1 g/kg, 2 g/kg and 4 g/kg were (2.23%, -5.39%, -10.43%) respectively. At 2 hour after subcutaneous injection of adrenaline, mean percent reductions of hyperglycaemia with 70% ethanolic extract of *Mirabilis jalapa* L. plant at the doses of 1 g/kg, 2 g/kg and 4 g/kg were (6.35%, 3.92%, 13.45%) respectively. At 3 hour after subcutaneous injection of adrenaline, mean percent reductions of hyperglycaemia with 70% ethanolic extract of *Mirabilis jalapa* L. plant at the dose of 1 g/kg, 2 g/kg and 4 g/kg were (8.95%, 21.24%, 28.73%) respectively. At 4 hour after subcutaneous injection of adrenaline, mean percent reductions of hyperglycaemia with 70% ethanolic extract of *Mirabilis jalapa* L. plant at the doses of 1 g/kg, 2 g/kg and 4 g/kg were (14.94%, 26.18%, 31.64%) respectively. The mean percent reductions of hyperglycaemia with glibenclamide at 1 hour, 2 hour, 3 hour and 4 hour after subcutaneous injection of adrenaline were (33.91%, 33.99%, 47.89% and 60.59%) respectively.

In the comparison between hypoglycaemic effect 70% ethanolic extract of *Mirabilis jalapa* L. plant and glibenclamide, glibenclamide had more hypoglycaemic effect than 70% ethanolic extract of *Mirabilis jalapa* L. plant.

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References

Atmakuri, L. R. and S. Dath. (2010). **Current trends in herbal medicines.** Journal of Pharm Res. Vol. 3(1): 109-111.

Bhattacharyya, B. (1998). **Flowering Plants Taxonomy and Phylogeny.** Narosa Publishing House. New Delhi Madras Bombay Calcutta, London.

Chen, Y., S. L. Jothy, Z. Zakaria, Y. L. Lau, L. Y. Latha and S. Sasidharan. (2011). **Acute Oral Toxicity of Methanolic Seed Extract of *Cassia fistula* in Mice.** Institute for Research in Molecular Medicine (INFORMM), Universiti Sains Malaysia.

Cooke, C. I. E. (1958). **The Flora of the Presidency of Bombay.** Vol. II. Printed by P.C. Ray, Sri Gouranga Press Private Ltd.,5, Chintamain Das Lane, Calcutta-9.

Cronquist, A. (1981). **An Integrated system of Classification of Flowering Plants,** Columbia University press New York.

Doss, V. A., R. Sowndarya and N. Moorthi. (2015). **Antidiabetic Activity of Hydroethanolic Extracts of *Mirabilis jalapa* L. Leaves in Streptozotocin Induced Diabetic Rats.** International journal of pharmacy and pharmaceutical research. Volume 4, Issue: 2.

Gupta, D. (2012). **Study of acute, Sub acute and chronic toxicity test.** International Journal of Advanced Research in Pharmaceutical and Bio Sciences. Vol. 2 (2): 103-129.

Gupta, S. S., S. C. L. Verma, V. P. Gerg, and M. Ri, (1967). **Antidiabetic effect of *Tinospora cordifolia.*** India Journal of Medicinal Research, 545 (7): 733-745.

Marles, R. J. and N. R. Farnsworth. (1995). **Antidiabetic plants and their active constituents.** Phytomedicine 2(2): 137-189.

Pallab, K., D. Satyendra and S. R. Kanta. (2014). **A review on ethnic plants of assam (India) and their anti-oxidant activity.** Journal of Medical Pharmaceutical And Allied Sciences. Vol-3. Institute of pharmacy, down town university, Guwahati Assam, India.

Pandey, M., M. Debnath, S. Gupta and S. K. Chikara. (2011). **Phytomedicine: An ancient approach turning into future potential source of therapeutics.** Journal of Pharmacognosy and Phytotherapy. Vol. 3, March.

Prakash, R. S., K. Madhab and S. Rosalin. (2012). **Antihyperglycaemic Activity of Aerial parts of *Mirabilis ja*lapa L.** In Normoglycemic and Streptozotocin-induced Hyperglycemic Rats. International journal of drug development and research. Vol. 4, Issue 4.

Sarkar, P., A. K. Mahmud and J. P. Mohanty. (2011). **Antidiabetic Activity of Ethanolic Extract of *Mirabilis jalapa* L. Root.** International journal of pharmacy and technology. Vol. 3, Issue No. 1: 1470-1479.

OECD 423. (2001). **Guidelines for the Testing of Chemicals Acute Oral Toxicity.** Acute Toxicity Class Method 423: 1-14.

World Health Organization. 2016. **Global Report on Diabetes.** Printed in France.