

# Effects of methanolic and aqueous extracts of *Ipomoea batatas* L on mineral contents level (calcium and magnesium) in alloxan-induced diabetic rats

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**Abstract:** In diabetic patients, electrolyte disorders frequently occur with the characteristic changes in minerals like calcium and magnesium etc. Several medicines are used to manage diabetes mellitus but they exert adverse effects. Plants are a valuable alternative to synthetic medicines because they are easily available, economical and have fewer side effects. *Ipomoea batatas* L is a well-known antidiabetic plant (sweet potato) but its effects on calcium and magnesium concentration have not studied. The prime focus of this study is to estimate the potential of *Ipomoea batatas* L peel-off on magnesium and calcium level in Alloxan-induced diabetic rats. Alloxan monohydrate was mixed in 0.9% NaCl solution and administrated [150 mg/kg (S/C)] to male Wistar rats to induce diabetes. After three days blood samples were collected and blood glucose level was recorded. Wistar rats having a blood glucose level of 200 mg/dl and above were selected for the study. Methanol and water extract of *Ipomoea batatas* L peel-off was given orally with a dose rate of 4g/day. Calcium and magnesium estimation was done using an atomic absorption spectrophotometer. Our results revealed an increase in both the calcium and magnesium level in heart, brain, liver, hind limb, and forelimb after *Ipomoea batatas* extract treatment. In kidneys decreased calcium level was noted as they excrete calcium. Mineral (Calcium, magnesium) level was increased in all organs except kidney after both extracts treatment. *Ipomoea batatas* being anti-diabetic in nature also maintain the homeostasis of calcium and magnesium in diabetes. Therefore, we propose the long-term use of such agents might help in the prevention of diabetes-associated complications. However, the validation of these results to human population needs further extensive study.

**Keywords:** *Ipomoea batatas*, calcium, magnesium, and alloxan-induced diabetes.

## INTRODUCTION

Non-insulin dependent diabetes mellitus (type II) and insulin dependent diabetes mellitus (type I) are the two major categories of diabetes mellitus (Surya *et al.*, 2014).

Global incidence of diabetes in adults in 1995 was anticipated to be 4% and elevated to 5.4% by the year 2025. In Europe and North America, 2 million people were affected by this disease (Hyoty, 2002).

Diabetes, especially type-2 diabetes is basically associated with the diet as a diet of the individuals play an important role in the prevention and treatment of diabetes. There are many available foods with known antidiabetic activity and *Ipomoea batatas* is one of them (Gunn *et al.*, 2013). *Ipomoea batatas* (sweet potato) a widespread and is eaten as a food in many countries like Pakistan, India, Japan, New Zealand etc and is a rich source of a number

of minerals, vitamins as well as other components. *Ipomoea batatas* contain a very good antidiabetic activity (Ijaola *et al.*, 2014) and used traditionally for the treatment of diabetes in various countries like Philippines (Gunn *et al.*, 2013). *Ipomoea batatas* decrease the blood glucose level significantly. *Ipomoea batatas* decrease the blood glucose level by increasing the insulin sensitivity rather than insulin secretion (Ludvik *et al.*, 2002).

In many research studies, a relationship between trace elements and diabetes mellitus was observed. In different studies an alteration in the metabolism of these trace elements (calcium and magnesium were noted (Hans *et al.*, 2002). Calcium and magnesium enhance the action of insulin by activating insulin receptor. They serve as a cofactor for various enzymes of glucose metabolism. There is an association between glucose intolerance and serum calcium level (Pittas *et al.*, 2007).

Magnesium has a critical role in the phosphorylation reaction of glucose and its metabolism. In diabetes, its

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deficiency occurred due to insulin resistance and increased urinary losses. Glucose-transporting mechanism of the cell membrane and various enzymes in carbohydrate oxidation involve magnesium as a co-factor. Magnesium is also involved in insulin secretion, insulin binding and activity. Magnesium is significant to diverse processes critical to glucose metabolism, cardiovascular functions and insulin action. In diabetic and cardiovascular disorders magnesium deficiency was observed (Hans *et al.*, 2002).

Besides anti-diabetic activity, the *Ipomoea batatas* contains a number of minerals as sodium, potassium, calcium, iron, magnesium, zinc, copper, and phosphorus (Montejo *et al.*, 2015). As the low levels of these minerals are linked with the development of diabetes and its complications, so the *Ipomoea batatas* plays a critical role in maintaining and hence controlling diabetes. Hypomagnesaemia is directly related with micro and macrovascular complications e.g. cardiovascular, retinopathy and nephropathy (Sales and Pedrosa, 2006). Diabetic patients have an inclination to develop magnesium deficiency. The plasma magnesium concentration is inversely correlated with insulin sensitivity. Magnesium supplementation increase insulin sensitivity and also insulin discharge in type-2 diabetic patients (Valk, 1999). Magnesium deficiency results in carbohydrate intolerance, insulin resistance and dyslipidemia. Magnesium deficiency increases diabetic complications, i.e. ischemic heart disease and retinopathy (Resnick *et al.*, 1991).

Calcium ion plays an important role in glycemic control by affecting the biosynthesis and release of insulin from *Beta cells* of the pancreas. There is a significant correlation between diabetes and hypocalcemia (Al-Yaseen *et al.*, 2009). Low serum ionized calcium concentration was the important factor in insulin-dependent diabetes. Decreased serum magnesium and calcium ions concentration were observed in the case of diabetes mellitus. Decrease calcium level relates to etiology and pathogenesis of diabetes mellitus. It is also reported that combined high intake of vitamin D and calcium is inversely associated with risk of incident type 2 diabetes mellitus (Anastassios *et al.*, 2007).

However appropriate trace element is beneficial in ameliorating some physiological deficiencies linked with diabetes mellitus and prevent secondary complications (Tuvemo and Gebre-Medhin, 1983). Hence it is essential to discover and advance such alternative source of medicine which would be cost-effective and have better efficacy with fewer or no side effects and help to prevent complication. Previous studies conducted by Willcox *et al.*, 2009; Miyazaki *et al.*, 2005; Islam, 2006 indicate that *Ipomoea batatas* has anti-diabetic potential. However, its effect on calcium and magnesium concentration has been indicated by a few studies. Therefore, in the present study,

we have tried to find out its effects on calcium and magnesium in alloxan-induced diabetic mice as a way of managing diabetes.

## **MATERIALS AND METHODS**

### ***Preparation of plant extract***

*Ipomoea batatas* was purchased from local market of Faisalabad (Pakistan). The plant was authenticated and the specimen was kept in the herbarium of pharmacy department GC University, Faisalabad. Roots of *Ipomoea batatas* were used in the current study. Roots of *Ipomoea batatas* were washed with distilled water in order to remove any external material or dust. Chopping of plant roots was done using chopper and roots were powdered. Maceration process was used for the extraction process. Powdered root (1000 g) was macerated in methanol (2000 ml) for the extraction purpose. Likewise, powder root (1000 g) was macerated in distilled water (2000 ml) for the extraction purpose separately. During the extraction, process extract was shaken intermittently (3-4 times a day). Extraction was performed for 7 days using the maceration process. The plant was successively macerated for a total of 21 days. After each extraction, extracts were filtered by Whatman filter paper no 1 and evaporated to dryness via rotary evaporator under reduced pressure. The semisolid extracts were dried using a water bath. Dried extracts were weighed and stored at room temperature in a well-closed inert container for further examination.

### ***Chemicals and drugs***

Glibenclamide (10 mg/kg) was purchased from the local market. Other chemicals and drugs e.g. Alloxan (ALX 90.5% w/v in Tween 80 solution) and NaCl were obtained from Merck Germany. Standard solution of calcium and magnesium were of analytical grade. All the solvents used were of analytical grade.

### ***Experimental animals***

Normal male Wistar rats (Laboratory bred) were used in the present study. Animals were acclimatized for 14 days. Animals were kept under standard animal housing conditions. Throughout the study, animals were provided with unlimited access to standard diet and water. The study was undertaken with due approval by the Institution Animal Ethics Committee. The ethical committee investigates and monitors the housing of animals and ensures that it is as per standards. This committee also ensures that animals used for the experiment are accurately handled prior and after the experiment. The ethical committee also ensures that records are properly maintained with respect to the experiment performed on animals. This committee also ensures that experiments are not performed simply for the intention of achieving manual skills. This committee organizes an in-house training program for any animal experiment to ensure quality research and welfare of animals (Balls 1994).

### **Induction of diabetes**

Diabetes was induced by administration of alloxan monohydrate [150 mg/kg (S/C)] after an overnight fasting for 12 hours (had to access only to water), to make them more prone to develop diabetes. Alloxan monohydrate was dissolved in (0.9% NaCl sol) and administered to male Wistar rats. After 3 days of alloxan monohydrate induction glucose level (GL) was monitored. Wistar rats having blood glucose level of 200 mg/dl and above after 3 days were selected for the current study. Blood glucose level was monitored using Blood glucose test diagnostic strips (NIPRO Blood diagnostic strips). Blood glucose was determined by amputation of tail tip under mild anesthesia using NIPRO blood diagnostic strips. All rats possessing glucose level less than 200 mg/dl were excluded from the current study.

### **Experimental group**

The diet of total 40 rats was considered standard and consisted of broiler ration No. 13. The animals were kept at standard housing conditions with 12 hours light/ dark cycle. These rats were kept in an animal house situated at the department of physiology and pharmacology, University of Agriculture, Faisalabad. Animals were kept at water *ad libitum* and standard diet. Treated groups were exposed orally to water extract and methanol extract with a dose rate of 4g/Kg/day daily. The animals were divided into 8 groups and a total number of rats were 48. Blood glucose level (BGL) was measured for all 8 groups and for each rat separately and recorded. Blood glucose level and body weight of experimental rats were recorded before starting the dose and at 3<sup>rd</sup>, 6<sup>th</sup>, 9<sup>th</sup>, 12<sup>th</sup> and 15<sup>th</sup> day of the experiment.

A total number of rats were 48. The rats were divided into 8 groups. Each group contains 6 animals.

Group 1: Normal control young animals

Group 2: Normal control old animals

Group 3: Diabetic control young animals

Group 4: Diabetic control old animals

Group 5: Aqueous extract treated young diabetic rats

Group 6: Aqueous extract treated old diabetic rats

Group 7: Methanol extract treated young diabetic rats

Group 8: Methanol extract treated old diabetic rats

Aqueous and methanolic extracts (4g/kg/day) were administered orally. After completion of experiment (14 days) blood of rats were taken. Blood samples were withdrawn from all the respective organs e.g. liver, kidney, lungs, heart, forelimbs and hind limbs.

### **Physical parameters**

Body weight (fig. 1), feed consumption and water intake of each rat were measured daily at 9:00 AM.

### **Body weight (g)**

Mean body weight (g  $\pm$ SE) of young and old rats at various days in normal and diabetic groups treated with water extract and methanol extract of sweet potato peel-off is tabulated in figs. 1 & 2 respectively. This interaction

was not significant, while the interaction of overall mean of body weight with days was significantly different. The highest body weight was shown on the 7<sup>th</sup> day, while the overall of lowest body weight was shown by the rats on the 12<sup>th</sup> day. The overall mean of highest body weight was shown by the old control group rats and the lowest overall mean of body weight was given by the young rats of diabetic treated groups.

### **Sample collection**

Rats of all the groups were decapitated for the collection of blood samples in heparinized tubes. Plasma was separated after centrifugation and stored in small aliquots at -4°C for further analysis

### **Sample interpretation**

After 14 days rats were killed by cervical dislocation and their abdomen is wide opened. A blood sample was withdrawn from all the respected organs e.g. liver, kidney, brain, heart, lung, forelimbs and hind limbs respectively.

The blood samples were analyzed using Atomic-Absorption Spectrophotometer (Perkin-Elmerizers). The standard stock solution of calcium (Ca) and magnesium (Mg) were prepared using standard conditions and calibrated. There Ca and Mg level was determined for both water and methanolic extracts separately. Qualitative analysis is performed for the presence of Ca and Mg in all the respective organs (liver, kidney, brain, heart, lung, forelimbs and hind limbs).

## **STATISTICAL ANALYSIS**

Mean value is calculated for both methanolic and aqueous extract treated diabetic rats. Mean was calculated using R software for all the experimental groups. Value <0.05 were considered significant.

## **RESULTS**

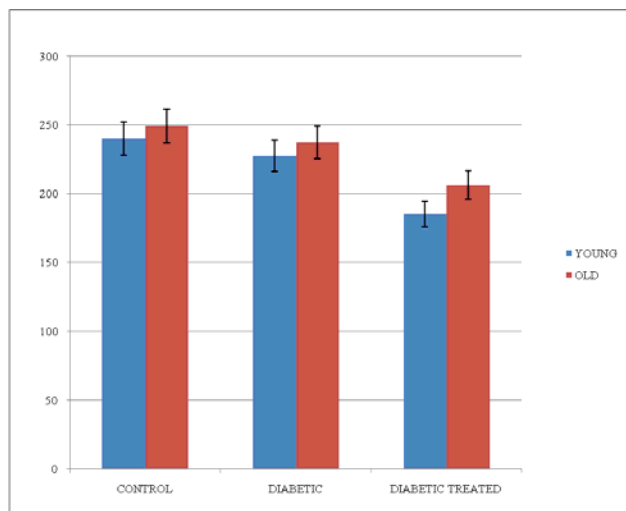
### **Effect of Water and methanol extract of *Ipomoea batatas* on Electrolytes and trace elements**

Results indicate that both aqueous and methanolic extract of *Ipomoea batatas* increases the calcium and magnesium level after treatment.

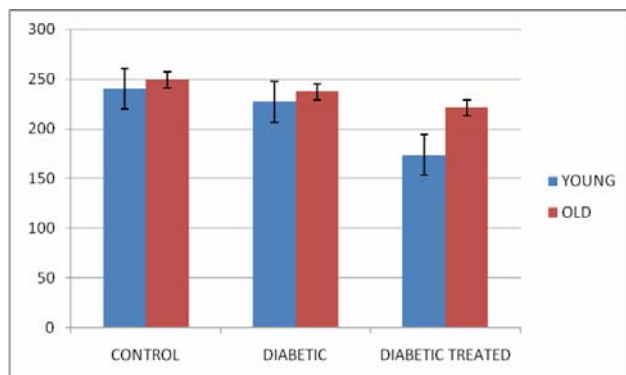
### **Calcium (Ca)**

*Liver, kidney, heart, brain, lungs, Forelimb, Hind limb (mg/100g)*

Mean calcium liver, kidney, heart, brain, lungs, forelimb and hindlimb level of old and young age rats of all treatment groups (control, diabetic, methanol and aqueous) are tabulated in table 1. After treatment with both water extract and methanol extract, the calcium level was increased in both diabetic young and old rat groups. The calcium level in liver and hindbrain was slightly higher with methanol extract treatment than water extract treatment. However, in all other organs, the increase in calcium level was similar and varied non-significantly with both treatments.



**Fig. 1:** Mean body weight (g ± SE) of young and old rats in normal control, diabetic and diabetic groups treated with water extract of sweet potato peel-off.



**Fig. 2:** Mean body weight (g ± SE) of young and old rats in normal control, diabetic and diabetic groups treated with methanol extract of sweet potato peel-off.

### Magnesium (Mg)

*Liver, kidney, heart, brain, lungs, Forelimb, Hind limb (mg/100g)*

Mean magnesium liver, kidney, heart, brain, lungs, forelimb and hindlimb level of old and young age rats of all treatment groups (control, diabetic, methanol and aqueous) are tabulated in table 2. After treatment with both water extract and methanol extract, the magnesium level was increased in both diabetic young and old rat groups. However, in rats with methanol extract treatment showed the higher increase in their magnesium level profile in almost in heart and hind limb as compared with water extract treated rats while in all other organs the increase was similar and varied non-significantly.

## DISCUSSION

Currently, the diabetes is approximately become a severe public health problem predominantly in developing countries (Deepa *et al.*, 2013). A variety of management including natural products, dietary supplements and

synthetic medicines are used to manage diabetes and related complications (Trojan *et al.*, 2012). Plants are the imperative basis of medicinal uses and potential bioactive constituents for the development of novel chemotherapeutic agents (Umamehesh and Ciddi., 2015).

Alloxan is the most extensively used drug for the induction of diabetes in the investigational animal model (Rohilla and Ali, 2012). It leads to a decrease in plasma insulin concentration leading to a constant hyperglycemic state. Hyperglycemia was observed after 3 days of Alloxan induction (Raju *et al.*, 2012). Alloxan induced diabetes constantly created the chief characteristics of diabetes mellitus including polydipsia, polyphagia, polyuria, decreases insulin level, weight loss and hyperglycemia. The standard drug glibenclamide acts by promoting insulin release by shutting of potassium-ATP channels, membrane depolarization and stimulation of calcium entry, a primary footstep in insulin emission (Goyal *et al.*, 2010). Calcium level was increased after treatment and increase in Ca level was helpful in controlling the diabetes level in alloxan-induced diabetic rats as calcium ion plays an important role in glycemic control by affecting the biosynthesis and release of insulin from *Beta cells* of the pancreas (Al-Yaseen *et al.*, 2009). There is a significant correlation between diabetes and hypocalcemia (Govindappa *et al.*, 2015). Altered calcium and vitamin D homeostasis plays an important role in the development of diabetes. Disturbance in calcium and vitamin D homeostasis results in insulin resistance, diabetes, metabolic syndrome and glucose intolerance. It is also reported that combined high intake of vitamin D and calcium is inversely associated with risk of incident type 2 diabetes mellitus (Anastassios *et al.*, 2007). Magnesium has a critical role in the phosphorylation reaction of glucose and its metabolism. Magnesium plays an essential role as a co-factor in countless enzymatic reactions involving energy metabolism. Glucose-transporting mechanism of the cell membrane and various enzymes in carbohydrate oxidation involve magnesium as a co-factor. Magnesium is also involved in insulin secretion, insulin binding and activity. Magnesium is significant to diverse processes critical to glucose metabolism, cardiovascular functions and insulin action (Sales and Pedrosa 2006).

With the treatment of water extract and methanol extract of *Ipomoea batatas*, the young and old rats showed the significant increase of magnesium level in the almost all the body organs. This increase in the magnesium level may lead to the control of hyperglycemia in diabetic rats of both groups as magnesium is linked with the control of glucose level (Valk, 1999).

Magnesium deficiency results in carbohydrate intolerance, insulin resistance and dyslipidemia. Magnesium deficiency increases diabetic complications, i.e. ischemic heart disease and retinopathy (Resnick *et al.*,

**Table 1:** Mean values of calcium in liver, kidney, heart, brain, lungs, fore limb and hind limb level of old and young age rats of all treatment groups

Organs ↓	Group	Control	Diabetic	Aqueous extract	Methanolic extract	Overall mean
Liver	Young	3.62±0.02 <sup>f</sup>	5.19±0.03 <sup>de</sup>	4.35±0.05 <sup>ef</sup>	5.90±0.01 <sup>cd</sup>	4.78±0.03 <sup>B</sup>
	Old	3.62±0.01 <sup>f</sup>	6.92±0.01 <sup>bc</sup>	7.36±0.02 <sup>b</sup>	11.47±0.04 <sup>a</sup>	7.34±0.02 <sup>A</sup>
	Overall mean	3.62±0.01 <sup>C</sup>	6.06±0.02 <sup>B</sup>	5.86±0.07 <sup>B</sup>	8.70±0.08 <sup>A</sup>	6.06±0.30
Kidney	Young	3.80±0.01 <sup>e</sup>	3.31±0.02 <sup>e</sup>	4.62±0.03 <sup>de</sup>	5.81±0.05 <sup>cd</sup>	4.39±0.05 <sup>B</sup>
	Old	4.16±0.03 <sup>de</sup>	7.70±0.04 <sup>ab</sup>	6.96±0.04 <sup>bc</sup>	8.82±0.01 <sup>a</sup>	6.89±0.01 <sup>A</sup>
	Overall mean	3.98±0.01 <sup>C</sup>	5.50±0.01 <sup>B</sup>	5.76±0.01 <sup>B</sup>	7.31±0.21 <sup>A</sup>	5.64±0.30
Brain	Young	3.74±0.34	5.25±0.65	6.27±0.50	6.53±0.55	5.45±0.35
	Old	3.74±0.34	4.94±0.62	4.34±0.23	6.45±0.18	4.86±0.29
	Overall mean	3.74±0.23 <sup>C</sup>	5.08±0.43 <sup>B</sup>	5.37±0.41 <sup>B</sup>	6.49±0.27 <sup>A</sup>	5.15±0.23
Heart	Young	4.53±0.30	5.94±0.06	6.20±0.20	6.76±0.58	5.86±0.25 <sup>B</sup>
	Old	4.53±0.30	5.88±0.76	7.55±0.18	8.54±0.46	6.63±0.42 <sup>A</sup>
	Overall mean	4.53±0.20 <sup>C</sup>	5.91±0.36 <sup>B</sup>	6.88±0.26 <sup>A</sup>	7.65±0.46 <sup>A</sup>	6.24±0.24
Lung	Young	4.63±0.23 <sup>bc</sup>	6.69±0.72 <sup>a</sup>	7.80±0.11 <sup>a</sup>	7.74±0.51 <sup>a</sup>	6.72±0.36 <sup>A</sup>
	Old	4.63±0.23 <sup>bc</sup>	3.92±0.63 <sup>c</sup>	5.22±0.38 <sup>bc</sup>	5.30±0.17 <sup>b</sup>	4.77±0.22 <sup>B</sup>
	Overall mean	4.62±0.16 <sup>B</sup>	5.30±0.65 <sup>B</sup>	6.51±0.47 <sup>A</sup>	6.52±0.48 <sup>A</sup>	5.74±0.26
Forelimb	Young	4.81±0.76	4.53±0.63	4.42±0.33	5.21±0.63	4.74±0.28 <sup>B</sup>
	Old	5.65±0.60	6.98±0.94	7.84±0.34	6.08±0.33	6.64±0.34 <sup>A</sup>
	Overall mean	5.23±0.48	5.75±0.67	6.13±0.61	5.65±0.33	5.70±0.26
Hind limb	Young	4.53±0.44 <sup>cd</sup>	3.85±0.22 <sup>d</sup>	5.93±0.64 <sup>b</sup>	7.48±0.32 <sup>a</sup>	5.45±0.36 <sup>A</sup>
	Old	4.52±0.45 <sup>cd</sup>	4.78±0.16 <sup>cd</sup>	4.39±0.28 <sup>cd</sup>	5.33±0.33 <sup>bc</sup>	4.75±1.70 <sup>B</sup>
	Overall mean	4.53±0.30 <sup>BC</sup>	4.31±0.21 <sup>C</sup>	5.16±0.33 <sup>B</sup>	6.40±0.42 <sup>A</sup>	5.10±5.10

**Table 2:** Mean values of magnesium in liver, kidney, heart, brain, lungs, fore limb and hind limb level of old and young age rats of all treatment groups

Organs ↓	Group	Control	Diabetic	Aqueous extract	Methanolic extract	Overall mean
Liver	Young	14.91±0.35 <sup>d</sup>	12.05±0.80 <sup>ef</sup>	12.99±0.62 <sup>de</sup>	10.66±0.31 <sup>f</sup>	12.65±0.44 <sup>B</sup>
	Old	35.98±0.54 <sup>a</sup>	29.36±0.21 <sup>b</sup>	23.31±0.92 <sup>c</sup>	23.17±0.90 <sup>c</sup>	27.96±1.28 <sup>A</sup>
	Overall mean	25.45±3.53 <sup>A</sup>	26.71±2.98 <sup>B</sup>	18.15±1.80 <sup>C</sup>	16.92±2.13 <sup>C</sup>	20.31±1.40
Kidney	Young	9.87±0.69 <sup>f</sup>	12.32±0.88 <sup>e</sup>	8.71±0.26 <sup>f</sup>	9.74±0.16 <sup>ef</sup>	10.16±0.40 <sup>B</sup>
	Old	29.21±0.55 <sup>a</sup>	25.65±0.54 <sup>b</sup>	18.20±0.72 <sup>d</sup>	21.46±0.74 <sup>c</sup>	23.63±1.00 <sup>A</sup>
	Overall mean	19.54±3.25 <sup>A</sup>	18.98±2.28 <sup>A</sup>	13.45±1.62 <sup>C</sup>	15.60±1.99 <sup>B</sup>	16.90±1.20
Brain	Young	14.10±0.62	14.61±1.82	10.36±0.37	10.20±0.21	12.32±0.65 <sup>B</sup>
	Old	28.04±2.00	26.66±1.74	18.84±0.59	18.66±0.54	23.04±1.18 <sup>A</sup>
	Overall mean	21.07±2.54 <sup>A</sup>	20.64±2.33 <sup>A</sup>	14.60±1.44 <sup>B</sup>	14.43±1.44 <sup>B</sup>	17.68±1.09
Heart	Young	15.87±1.33	11.18±0.77	11.35±0.65	23.09±13.70	15.37±3.36 <sup>B</sup>
	Old	30.35±2.73	27.85±0.56	19.09±0.69	20.44±0.50	24.43±1.28 <sup>A</sup>
	Overall mean	23.11±2.81	19.52±2.81	15.22±1.37	21.77±6.48	19.90±1.92
Lung	Young	13.87±1.81 <sup>d</sup>	11.63±0.85 <sup>de</sup>	12.96±0.39 <sup>d</sup>	9.65±0.33 <sup>e</sup>	12.03±0.60 <sup>B</sup>
	Old	30.92±0.87 <sup>a</sup>	23.17±1.73 <sup>b</sup>	19.60±0.81 <sup>c</sup>	20.33±0.58 <sup>bc</sup>	23.50±1.14 <sup>A</sup>
	Overall mean	22.40±3.00 <sup>A</sup>	17.40±2.13 <sup>B</sup>	16.28±1.18 <sup>BC</sup>	14.50±1.81 <sup>C</sup>	17.77±1.12
Fore-limb	Young	13.49±1.76	13.21±1.39	13.70±1.16	18.72±0.48	14.78±0.79 <sup>B</sup>
	Old	33.43±2.11	33.71±0.57	27.20±0.86	23.50±1.08	29.46±1.15 <sup>A</sup>
	Overall mean	23.46±3.57 <sup>A</sup>	23.46±3.49 <sup>A</sup>	20.45±2.3 <sup>5A</sup>	21.11±0.97 <sup>AB</sup>	22.12±1.36
Hind-Limb	Young	14.82±1.26	11.53±1.08	13.90±0.51	9.19±0.17	12.36±0.64 <sup>B</sup>
	Old	37.78±1.23	29.51±0.92	25.16±1.74	32.22±0.88	31.17±1.19 <sup>A</sup>
	Overall mean	26.30±3.92 <sup>A</sup>	20.52±3.07 <sup>B</sup>	19.53±3.06 <sup>B</sup>	20.71±3.88 <sup>B</sup>	21.76±1.65

1991). Hypomagnesaemia is directly related with micro and macrovascular complications e.g. cardiovascular, retinopathy and nephropathy. In the case of diabetes mellitus, its deficiency occurred due to insulin resistance and increased urinary loses. Magnesium supplementation

increase insulin sensitivity and also insulin discharge in type-2 diabetic patients (Valk, 1999).

Minerals such as calcium and magnesium levels are decreased in the case of diabetes. The decrease in these

mineral levels causes further complications of diabetes. Increase in this mineral level may be a key factor in the treatment of diabetes mellitus. Increase in these mineral level leads to an increase in insulin level which decreases glucose level in the body. So the plants which increase the mineral level can be used in the treatment of diabetes. There are four probable explanations for the antidiabetic activity of *Ipomoea batatas*. Initially, in pancreatic *Beta cells*, *Ipomoea batatas* modulate the calcium homeostasis. Disturbances in calcium homeostasis increase the diabetic related complications. *Ipomoea batatas* is rich in minerals (calcium), so this plant modulates calcium homeostasis. Secondly, this plant also increases magnesium concentration in alloxan-induced diabetic rats. Magnesium is necessary for insulin secretion. As *Ipomoea batatas* is rich in minerals (magnesium) so it increases insulin release. Basically, *Ipomoea batatas* is secretagogues in nature as it increases insulin release from *Beta cells* of Pancreas. Thirdly, *Ipomoea batatas* also restore DNA breakage induced by alloxan. Finally, it may have produced its effect by preventing the fatality of *Beta-cells*. *Ipomoea batatas* also permit the recovery of partly damaged *B-cells*, so this plant may have preliminary cell proliferation. In our current study, the diabetic effect of an aqueous and methanolic extract of *Ipomoea batatas* was observed on male Wistar rats. The results indicate that both water and methanolic extract shows potent *anti-diabetic* activity. Furthermore, methanolic extract shows more promising results in normalizing blood glucose level as compared to aqueous extract. *Ipomoea batatas* normalize blood glucose level in alloxan-induced diabetic Wistar rats. The result of the study clearly indicates the *anti-diabetic* activity of this medicinal plant. Apart from *anti-diabetic* activity the plant also shows the promising effect on mineral contents. *Ipomoea batatas* increase the mineral level (calcium and magnesium) in alloxan-induced diabetic rats. Mineral level such as calcium and magnesium level decreases in diabetes mellitus. This plant shows potent anti-diabetic activity and also increase mineral level in alloxan-induced diabetic rats.

Aqueous and methanolic extract increase the mineral level in liver, heart, brain, lungs, forelimb and hind limb. Aqueous and methanolic extract decrease the calcium in kidneys. This is because kidneys excrete out the calcium from the body. In young rats, the overall mean of calcium level in liver, kidneys, forelimbs and heart was found to be lower non-significantly than old age rats [table1]. The reason for the decreased level in young was may be due to less water intake and low body weight than in older rats. The activity of aldosterone may be more pronounced than the young rats. The mean calcium level in diabetic treated rats of all given organs was non-significant. With both extracts that increased in calcium level that may be due to a high level of calcium in sweet potato which leads to increased level after treatment (Suda *et al.*, 2003).

Diabetes also leads to an increase in calcium level due to bone osteopenia may also contribute to that increased level of calcium level. Aqueous and methanolic extract increase the mineral level in liver, heart, brain, lungs, forelimb and hind limb. Aqueous and methanolic extract decrease the magnesium in kidneys. This is because kidneys excrete out the magnesium from the body. In young rats, the overall mean of magnesium level in liver, kidneys, heart and hind limb was found to be lower than old age rats. While in other remaining organs of diabetic treated rats given the opposite pattern of magnesium level than above. In old rats, the overall mean of magnesium level in all given organs were more than the young rats (table 2). The overall mean values of magnesium level in all given organs after treatment with both extracts of diabetic treated rats were less than their control group rats. This may be due to improvement in diabetes because in diabetes magnesium level was increased (Barbagallo and Dominguez, 2015). Secondly, the absorption of magnesium decreased in the presence of excess calcium which also leads to low magnesium level in diabetic treated rats.

## CONCLUSION

Minerals (calcium & magnesium) concentration decreases in the case of diabetes mellitus. *Ipomoea batatas* has the ability to enhance the level of magnesium and calcium in various organs of Alloxan-induced diabetic rats. *Ipomoea batatas* decreases diabetic complications by increasing calcium and magnesium level in different organs. There is a constant study to isolate and differentiate the bioactive compound responsible for this action. *Ipomoea batatas* is anti-diabetic in nature and possess the ability to enhance calcium and magnesium level. These two properties make this plant a potential source for the new diabetic drug. Therefore, we purpose that long-term use of such agents might be helpful in the prevention of diabetes-associated complications. However, to know the function of *Ipomoea batatas* for enhancing the trace elements like calcium and magnesium in diabetes, clinical trials are needed enrolling a large number of cases. Further studies are needed to clearly demonstrate the mode of action of this anti-diabetic.

## REFERENCES

- Al-Yaseen, Nimmim T and Challab D (2009). Calcium and diabetes mellitus type Two a prospective study done on people with type 2 diabetes in Diwaniya teaching hospital. *Kufa Med. J.*, **12**(1): 468-475.
- Anastassios GP, Harris SS, Stark PC and Hudes DB (2007). The effects of calcium and vitamin D supplementation on blood glucose and markers of inflammation in non-diabetic adults. *Diab. Care.*, **30**(4): 980-986.

- Balls M (1994). Replacement of animal procedures: Alternatives in research, education and testing. *Lab. Animals.*, **28**:193-211.
- Barbagallo M and Dominguez M (2015). Magnesium and type II diabetes. *World J. Diab.*, **6**: 1152-1154.
- Deepa SV, Rajaram K and Kumar PS (2013). *In vitro* and *in vivo* antidiabetic effect of *Andrographis lineata* Wall. Ex. Nees and *Andrographis serphyllifolia* Wt.lc leaf extract. *Afr. J. Pharm. Pharmacol.*, **7**(29): 2112-2121.
- Govindappa TS, Sadananda YL, Channabasave R, Chandrappa RS, Padmalatha SK and Prasard S (2015). *In vitro* and *in vivo* antidiabetic activity of lecithin ( N-acetyl-galactosaine, 64 kDs, isolated from endophytic fungi, Alternaria species from *Viscum album* on alloxan induced diabetic mice. *Integrat. Obes. Diabet.*, **1**(1): 11-19.
- Goyal S, Rai K, Narang K and Rajesh K (2010). Sulfonyl urease for anti-diabetic activity an overview for glipizide. *Inter. J. Pharm. Pharmaceut. Sci.*, **2**(2): 1-6.
- Gunn J, Che C and Farnsworth N (2013). Diabetes and natural products. *Bioact. Food Diet. Interven. Diab.*, **1**: 381-394.
- Hans P, Sialy R and Bansal D (2002). Magnesium deficiency and diabetes mellitus. *Curr. Sci.*, **83**: 12-25.
- Hyoty H (2002). Entero-virus infections and type 1 diabetes. *Ann. Med.*, **34**: 138-141.
- Ijaola T, Osunkiyesi A, Taiwo A, Oseni O, Lanrelyanda Y, Ajayi J and Oyede R (2014). Antidiabetic effect of *Ipomoea batatas* in normal and alloxan-induced diabetic rats. *IOSR J. Applied Chem.*, **7**(5): 16-25.
- Islam S (2006). Sweet potato (*Ipomoea batatas* L.) leaf: its potential effect on human health and nutrition. *J. Food Sci.*, **71**(2): 45-50.
- Ludvik BH, Mahdjoobian K, Waldhaeusl W, Hofer A, Prager R, Kautzky-Willer A and Pacini G (2002). The effect of *Ipomoea batatas* (Caiapo) on glucose metabolism and serum cholesterol in patients with type 2 diabetes: a randomized study. *Diab. Care*, **25**(1): 239-240.
- Miyazaki Y, Kusano S, Doi H and Aki O (2005). Effects on immune response of anti-diabetic ingredients from white-skinned sweet potato (*Ipomoea batatas* L). *Nutr.*, **21**(3): 358-362.
- Montejo JF, Mondonedo J, Lee M, Ples M and Vitor R (2015). Hematological effects of *Ipomoea batatas* (camote) and *Phyllanthus niruri* (sampa-sampalukan) from Philippines in the ICR mice (*Mus musculus*). *Asian Pac. J. Trop. Biomed.*, **5**(1): 29-33.
- Paolisso G, Scheen A, d'Onofrio F and Lefebvre P (1990). Magnesium and glucose homeostasis. *Diabetol.*, **33**(9): 511-514.
- Pittas A, Lau J, Hu B and Hughes D (2007). The role of vitamin-D and calcium in type-2 diabetes. A systemic review and meta-analysis. *J. Clin. Endocrinol. Metabol.*, **92**(6): 2017-2029.
- Raju S, Rao B and Latha M (2012). Antidiabetic activity of *Smilax China* roots in alloxan-induced diabetic rats. *Inter. J. Pharma. Tech. Res.*, **4**(1): 369-374.
- Resnick L, Gupta R, Bhargava K, Gruenspan H, Alderman M and Laragh J (1991). Cellular ions in hypertension, diabetes and obesity. A nuclear magnetic resonance spectroscopic study. *Hyperten.*, **17**(6): 951-957.
- Rohilla A and Ali S (2012). Alloxan induced diabetes: mechanism and effects. *Inter. J. Res. Pharmaceut. Biomed. Sci.*, **3**(2): 819-823.
- Sales H and Pedrosa L (2006). Magnesium and diabetes mellitus and their relation. *Clin. Nutr.*, **25**(4): 554-562.
- Suda I, Oki T, Masuda M, Kobayashi M and Nishiba Y (2003). Physiological functionality of purple-fleshed sweet potatoes containing anthocyanins and their utilization in food. *Japan Agri. Res. Quart.*, **37**: 167-173.
- Surya S, Salam A, Tomy D, Carla B, Kumar R and Sunil C (2014). Diabetes mellitus and medicinal plants-a review. *Asian Pac. J. Trop. Dis.*, **4**(5): 337-347.
- Trojan A, Rodrigues M, Alves T, Soares G and Ritter M (2012). Plants used as antidiabetics in popular medicine in Rio Grande do Sul, southern Brazil. *J. Ethnopharmacol.*, **139**(1): 155-163.
- Tuvemo T and Gebre-Medhin M (1983). The role of trace elements in juvenile diabetes mellitus. *Pediatr.*, **12**(4): 213-219.
- Umamehesh B and Ciddi V (2015). *In vivo* and *in vitro* evaluation of anti-diabetic and insulin secretagogue activities of *Capparis Zeylonica*. *Pharmacol. Pharm.*, **6**(1): 311-320.
- Valk H (1999). Magnesium and diabetes mellitus. *Netherlands J. Med.*, **54**(9): 139-146.
- Willcox D, Willcox B, Todoriki H and Suzuki M (2009). The Okinawa diet: health implications of a nutrient-dense, antioxidant-rich dietary pattern low in glycemic load. *J. Amer. Coll. Nutr.*, **28**(4): 500-551.