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# Glucose-6-phosphatase activity in selected rabbit tissues of normal and alloxan induced diabetic rabbit

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**ABSTRACT**

The effect of diabetes on glucose -6- phosphatase activity in selected tissues of rabbit was investigated. Rabbits were made diabetic by the administration of Alloxan (300mg/kg body weight) intraperitoneally. Fasting blood glucose level greater than 300mg/dl which persisted when untreated indicate Diabetes mellitus. Diabetic rabbits were treated with 100mg/kg body weight of aqueous extract from unripe pulp of Carica papaya. Result from normal and diabetic animals were compared with control administered distilled water only. In tissues of normal rabbit studied, liver, small intestine and stomach had a relative higher enzyme activity compared to the kidney. This is due to the fact that these organs especially liver and small intestine are highly involved in active transport processes and also useful for other purposes like regenerating glucose and ATP levels. Glucose-6-phosphatase activity increased significantly in the liver of diabetic animals. This shows that liver from diabetic rabbits exhibit increased glucose synthesis than normal animals which implies that Alloxan diabetes contributed to the elevation of this enzyme.

**Key words:** Alloxan diabetes, Alloxan, ATP and Glucose-6-Phosphatase.

## INTRODUCTION

Glucose-6-phosphatase is a $Mg^{2+}$ activated enzyme found in the luminal side of the endoplasm reticulum of hepatocytes and renal cells. The enzyme is not present in muscle or in the brain, and gluconeogenesis does not occur in these tissues, instead glucose produced by gluconeogenesis in the liver or kidney or ingested in the diet is delivered to brain and muscle through the blood stream. There have been many reports of glucose-6-phosphatase hydrolysis in other tissues i.e. small intestine (Guionie, *et al,* 2003). Glucose-6-phosphatase catalyzes the final step of glucose production by liver and kidney. Though its strategic position has sparked interest in its regulation. Previous work examining the physiologic regulation of this enzyme has relied on estimates of glucose-6-phosphatase activity in crude microsome preparation (Liu *et al*, 1994)

Glucose-6-phosphatase deficiency is a rare, inherited disorder of glycogen metabolism. This enzyme deficiency i.e. genetic deficiencies of any of the protein components of the glucose-6-phosphatase system are classified as glycogen storage disease (GSD) type 1a, also known as Von Gierke disease (Chen and Burchell, 1995). The clinical manifestation are due to the body's inability to breakdown glycogen into usable glucose molecules during periods of fasting, precipitating metabolic disarray and chronic counter regulatory (stress) hormone excesses. The pathophysiology of glucose-6-phosphatase deficiency and its consequences are complex, but they form the basis for understanding the clinical manifestation of the disease. Without glucose-6—phosphatase, glucose cannot be released from glycogen breakdown during periods of fasting. This caused fasting hypoglycaemia, the primary manifestation of this disorder (Baker *et al*, 1989; Goldberg and Slonum, 1993).

Another manifestation of the disease is that glucose-6-phosphatase and glycogen accumulate in the liver. This occurs because neither can be degraded, and no down-regulation of the pathways occurs that allows glycogen synthesis following a meal. This result in characteristic hepatomegaly due to glycogen excess within the hepatic cells. The pathophysiology of glucose-6-phosphatase deficiency accurately explains the clinical manifestations of hypoglycaemia, hepatomegaly, hyperlipidemia, hyperuricemia and poor growth and development (Fernandes et al, 1988; Greene et al, 1991).

Acute streptozotocin-diabetes increased expression of glucose-6-phosphatase mRNA and this contributes to the increased glucose-6-phosphatase activity seen with Diabetic mellitus. Hepatic microsomal glucose-6-phosphatase activity levels and the hepatic output of glucose are increased in diabetes (Guionie, *et al* 2003). The aim of this study is to monitor the pattern of Glucose-6-phosphatase activity in diabetic animals.

## MATERIAL AND METHOD

### Plant material

Fresh, unripe, mature fruits of *Carica papaya* were obtained from National Horticultural Research Institute (NIHORT) Ibadan, Nigeria. The fruits were peeled, seed removed and the pulp cut into small pieces, sun-dried and finely powdered with an electric grinder. The powdered material was stored in properly sealed bottles at $10^{o}$c in the refrigerator

### Determination of Protein

Protein concentration measurements were carried out conditions optimum for individual enzymes. All measurement were carried out using Spectroic 21 Spectrophometer, glass cuvettes of 1cm light path were used throughout. The concentration of serum and homogenates were determined using biuret method (Plummer, 1978)

### Measurement of glucose-6-phosphatase activity

Glucose-6-phosphatase activity was determined using the method described by Swanson (1950). The method is based on the incubation of the specific substrate with the enzyme and the determination of liberated inorganic orthophosphate.

## RESULT AND DISCUSSION

The activities of glucose-6-phosphatase in the selected tissues of normal as well diabetic rabbits following administration of aqueous extract of unripe pulp from *Carica Papaya* are shown in (Tables 1 & 2). No significant change was observed in the serum of normal and diabetic rabbits administered 100mg/kg body weight of the aqueous extract respectively. Other tissues with the exception of liver, demonstrated significant reduction in glucose-6-phosphatase activity.

In the tissues studied, liver, small intestine and stomach had a relatively higher enzyme activity compared to the kidney (Table 1). This is due to the fact that these organs especially liver and small instetine are highly involved in active transport

**Table 1: Effect of oral administration of aqueous extract of Carica papaya on Glucose-6-phosphatase (nM/pi/hr/mg.protein) in normal rabbit tissues***

| Group | Dose (mg/kg) | Serum | Small intestine | Stomach | Kidney | Liver |
|---|---|---|---|---|---|---|
| Normal untreated rabbits (control) | _ | $0.01\pm0.006^{a}$ | $0.02\pm0.00^{a}$ | $0.05\pm0.006^{a}$ | $0.02\pm0.01^{a}$ | $0.07\pm0.000^{a}$ |
| Normal | 50 | $0.01\pm0.00^{a}$ | $0.13\pm0.01^{b}$ | $0.13\pm0.01^{b}$ | $0.05\pm0.02^{b}$ | $0.25\pm0.06^{a}$ |
| treated | 100 | $0.02\pm0.00^{b}$ | $0.03\pm0.007^{c}$ | $0.04\pm0.007^{a}$ | $0.06\pm0.00^{c}$ | $0.007\pm0.02^{a}$ |
| rabbits | 200 | $0.02\pm0.01^{b}$ | $0.73\pm0.05^{d}$ | $0.27\pm0.03^{c}$ | $0.05\pm0.02^{b}$ | $0.95\pm0.2^{c}$ |

*Results are means of four determinations ± SEM. Values with different notations are statistically different ($p<0.05$)

processes. The liver endoplasmic reticulum glucose-6-phosphatase, catalyses glucose-6-phosphate hydrolysis during gluconeogenesis and glycogenolysis (Guyton and Hall, 2003). The high glucose-6-phosphatase observed in the liver and its presence in small instetine and stomach confirms the report of Guionie *et al,* (2003) that highest activities are found in the liver. The enzyme is located

**Table 2: Effect of oral administration of aqueous extract of Carica papaya on Glucose-6-phosphatase (nM/pi/hr/mg.protein) in normal rabbit tissues***

| Group | Dose (mg/kg) | Serum | Small intestine | Stomach | Kidney | Liver |
|---|---|---|---|---|---|---|
| Diabetic untreated rabbit | - | 0.02±0.01a | 0.18±0.04a | 0.15±0.1a | 0.10±0.03a | 0.13±0.01a |
| Diabetic treated rabbits | 100 | 0.02±0.01a | 0.11±0.04b | 0.13±0.04b | 0.05±0.04b | 0.67±0.04b |

*Results are means of four determinations ± SEM. Values with different notations are statistically different ($p<0.05$)

mainly with its catalytic site oriented towards the lumen of the endoplasmic reticulum in liver and kidney cells. The presence of glucose-6-phosphatase in tissue i.e. stomach, and small instetine, other than the liver, suggest its enzymatic activity might also be used for other purposes such as locally regenerating glucose stock or ATP levels (Ramos and de Meis, 1999).

The level of glucose-6-phosphatase is elevated in the liver and kidney of diabetic rabbits when compared with animals treated with *Carica papaya* fruit extract. This result obtained shows that glucose-6-phosphatase is an important enzyme in liver gluconeogenesis. Since glucose cannot be absorbed through peripheral tissues, liver and kidney depends on gluconeogenetic pathway for the production of glucose and ATP.

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