Z. Ernährungswiss. **17**, 79–83 (1978)

ISSN 0044–264 X

National Research Centre, Dokki, Cairo (Egypt),
and Kasr El-Aini Faculty of Medicine, Cairo University

Blood-reduced glutathione, serum ceruloplasmin and mineral changes in juvenile diabetes

R. Awadallah, E. A. El-Dessoukey, H. Doss, K. Khalifa, and *Z. El-Hawary*

With 1 table

(Received December 29, 1977)

Glutathione assumes a prime importance in carbohydrate metabolism due to its activation of the enzyme systems which affect carbohydrate metabolism, either directly or indirectly. These enzymes contain sulphydryl groups in their protein moiety, essential for the integrity of their action (20). The importance of the –SH groups in this system as well as in other enzyme systems was revealed to be due to the participation of the –SH in the active radical of CoA (29).

Zinc seems to be essential for the utilization of glucose (22), it enhances the hypoglycaemic action of insulin (8) and stimulates the uptake of glucose by rat epididymal fat (24).

Knjavez and *Yurkov* (14) found that in the absence of symptoms of decompensation the serum zinc concentration was low in juvenile diabetic patients, the decrease in zinc concentration was greater. Also, a significant increase of zinc excretion in urine was found in juvenile diabetic patients (7).

Vanghelovicia et al. (33) showed that there is a normal level of serum copper in diabetes, but that a highly significant inverse. Correlation exists between copper levels and the age of onset of symptoms of diabetes (2).

Calcium plays an important role in the mechanism of insulin release from the beta cells (25). It is hypothesized that calcium ions influx in the beta cell might trigger the release of insulin and that epinephrine might suppress insulin secretion by blocking such influx. *Josinki* et al. (11) found that total and ionized plasma calcium were decreased in diabetic patients.

Stutzman (30) found that patients requiring more than 20 units of NPH insulin had a significantly lower blood magnesium level than those with controlled diets. These findings agreed with that obtained by *Ruichi* et al. (26).

Kartelishev (12) reported that changes in the electrolyte pattern in juvenile diabetes especially during the phase of decompensation, but the pattern gradually returned to normal after insulin therapy. Increased level of potassium in plasma was observed, but in the erythrocytes the reverse occurred (12). Potassium levels tended to be higher than normal

on the first day of life in infants of mothers who were diabetic or in the prediabetic state (7).

The presence of hyponatermia was established in unstable juvenile diabetes (19). The situation appeared to be permanent, even in cases of compensated diabetes. *Kartelishev* (12) also reported a decrease in plasma sodium and an increase in erythrocyte sodium in diabetic children.

The present study was performed to study the metabolism of glutathione, ceruplasmin oxidase activity and certain inorganic elements such as zinc, copper, iron, calcium, magnesium, potassium and sodium in juvenile diabetic patients.

Material and methods

The subjects in this study were divided into two groups. The first group comprised twenty normal children selected on clinical and laboratory grounds, particular care was taken to exclude any liver, splenic affection or urinary and intestinal Bilharziasis, since large numbers of the local population are infected. The second group comprised 24 cases of established juvenile diabetes in the Student's Hospital Cairo.

Blood samples were taken from fasting subjects, a portion of the blood was used for analysis of glucose and reduced glutathione. The remaining portion was allowed to clot for two hours in polyethylene tubes before centrifugation and the serum was separated for the analysis of other pattern studies.

The method of *Nelson's* modification of *Somogyi's* (28) was used for determination of blood sugar. Blood-reduced glutathione was determined by the method of *Thompson* and *Watson* (32). The ceruloplasmin oxidase activity was determined by the method of *Henry* et al. (10). Serum zinc and copper were determined by the method of *Sinha* and *Gabriele* (27). Serum iron, potassium and sodium were estimated by the method published in *Beckman* Analytical method by Atomic Absorption Spectrophotometer. Serum calcium and magnesium were determined using the method of *Willis* (34).

Results and discussion

In our cases of juvenile diabetes, the level of the fasting blood-reduced glutathione was significantly lower than that of normal children, as shown in table 1. Our findings are in agreement with those of *Khattab* et al. (13) and *Lab* and *Kumar* (17). This significant decrease in blood-reduced glutathione level may be due to:

(I) Decreased synthesis of glutathione, this receives evidence from the work of *Krahl* (15), demonstrating incorporation of labelled glycine into glutathione in the liver slices of severely diabetic rats. Glucose was found to increase glutathione synthesis, while insulin restored it completely to normal. By analogy, it is possible that in human diabetes insulin deficiency and impaired glycolysis may be the cause of the decrease in blood-reduced glutathione.

(II) Increased requirements and rapid destruction of the –SH groups, this may be due to the abnormal formation of certain compounds which might interact with destroy the available glutathione sulphydryl groups. The formation of such compounds were suggested by *Lazarow* (18) and were thought to resemble alloxan and to be responsible for the development of diabetes.

Table 1. Fasting blood glucose, glutathione (GSH), serum ceruloplasmin activity, and serum mineral changes in controls and juvenile diabetes (mean ± S.D.).

Item	Normal controls	Juvenile diabetes	P >
Blood glucose mg%	100.95 ± 2.67	278.3 ± 20.29	0.005
Reduced GSH mg%	34.6 ± 0.86	21.8 ± 0.41	0.005
Serum zinc μg%	119.4 ± 3.93	70.1 ± 3.38	0.005
Serum copper μg%	105.8 ± 2.26	84.9 ± 2.97	0.05
Cerul. oxidase units	610.8 ± 80.48	550.3 ± 80.59	0.05
Serum iron μg%	102.2 ± 3.86	110.7 ± 5.18	*i.n.
Serum calcium mg%	9.8 ± 0.17	9.4 ± 0.17	i.n.
Serum magnesium mg%	3.7 ± 0.22	3.1 ± 0.22	i.n.
Serum potassium mg%	17.3 ± 0.39	16.9 ± 0.35	i.n.
Serum sodium mg%	324.7 ± 4.54	280.2 ± 3.99	0.025

*i.n.: Insignificant

Furthermore, *Lab* and *Kumar* (17) showed that blood-reduced glutathione was significantly low in diabetes with ketosis and suggested that the fall in blood glutathione in diabetes may be a consequence of disturbed electrolyte balance.

Serum zinc levels of diabetic children were significantly lower than that of normal children (70.1 ± 3.38 μg% as against 119.4 ± 3.93 μg%). Our results agreed with those obtained by *Krainick* et al. (16).

There has been no satisfactory explanation in the literature for the lower blood serum zinc of diabetes. Is it one of the causes of the diabetic syndrome, or is it a result?

We expected that the pancreatic juice is very rich in zinc and may be even richer than normal in diabetics. This may be one of the routes by which zinc is lost from the blood in diabetes, the other route is via the urine which is often excessive in the diabetic state. Some workers have reported that the urine of diabetic patients has a higher than normal concentration of zinc (5, 6).

Prasad et al. (23) have explained the lower serum zinc in many Egyptians by the presence of phytie acid in Egyptian bread which interfers with the absorption of zinc.

Another explanation, the hyperglycaemia occurring in diabetic children, may possibly be due at least in part to decreased glucose utilization by the tissues caused by a lower level of serum zinc.

The serum copper in our juvenile diabetic groups was slightly decreased (84.9 ± 2.97 μg%) as compared to normal children (105.8 ± 2.26 μg%).

With regard to the serum ceruloplasmin level, we found it to be 550.3 ± 80.59 units against 610.8 ± 80.48 units in normal children.

The medical literature on the serum copper and ceruloplasmin in juvenile diabetes is very scanty, and no exhaustive study of the correlation between serum copper and ceruloplasmin on the one hand and the diabetic state on the other.

The mean serum iron of our patients was 110.7 ± 5.18 μg%, when compared with normal children, there are no significant differences.

These findings are supported by those of *Heintzelmann* (9) who found also no change in the serum iron in diabetes.

There were normal levels of serum calcium and magnesium in our subjects of juvenile diabetes. Our findings are in agreement with those of *Poroa* (21) who found no significant difference in serum magnesium in compensated and non-compensated diabetes.

Svyatelik (31) observed that a disturbance in magnesium metabolism apparently affects cell membrane permeability in diabetes. Furthermore, it has been shown that magnesium stimulates the uptake of glucose by rat diaphragm in the presence of insulin. This is presumably because magnesium is a cofactor in many enzymes involved in carbohydrate metabolism such as intracellular ATP-ase.

In this work the mean serum potassium was 16.9 ± 0.35 mg%, when compared with normal children, there was no significant difference. *Alonso* (1) found that blood potassium was particularly decreased in subjects with a very high blood sugar. Other workers have reported, however, that in severe cases when signs of intracellular dehydration occur, the plasma potassium may be somewhat higher than normal (3). The explanation, why serum potassium is still normal in juvenile diabetes and the excessive loss of potassium in the urine, may be offset by the release of potassium into the blood as a result of the excessive transformation of muscle and tissue protein into carbohydrates, thereby releasing into the blood intracellular potassium.

The mean serum sodium was significantly lower from 324.7 ± 4.54 mg % in normal children to 280.2 ± 3.99 mg % in juvenile diabetes. Our findings are in concordance with those obtained by *Kartelishev* (12).

This is most probably due to the glucose-induced osmotic diuresis. Initially there are losses of water, sodium and chloride from the extracellular fluid, but if the glucosuria continues, losses also occur from the intracellular compartment (4). Therefore, in subjects with severe decompensated diabetes, there is a decrease in the sodium content of both the blood serum on the one hand and the erythrocytes and other body cells on the others in proportion to the severity of the disease.

Acknowledgement

Our thanks are due to Dr. *Nagib Zaki*, Head of the Diabetic clinic, Student's Hospital, for his great help and willing co-operation.

Summary

In cases of juvenile diabetes, there were lower than normal levels of reduced glutathione, ceruloplasmin oxidase activity, zinc, copper and sodium, while the other elements show no significant changes. The lower level of serum zinc, copper and sodium may be due to the osmotic diuresis and consequent polyurea of diabetes.

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Authors' address:
Dr. *Raafat Awadallah,* Biochemistry Department, National Research Centre, Dokki, Cairo (Egypt)