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Relationship Between Biochemical Parameters, Oxidative Stress Markers and Glycemic Control in Type II Diabetes Mellitus

Friday, October 11, 2019, 10:15 – 11:15 AM, 2:25 - 3:25 PM
Saturday, October 12, 2019, 10:00 – 11:00 AM, 2:15 - 3:15 PM

Fagbohun OF¹, Odewole TO², Emma-Okon BO³, Agboola FK², Kolawole BA⁴, Onakpoya OH⁵

1. Department of Biomedical Engineering, First Technical University, Ibadan, Oyo State, Nigeria; 2. Department of Biochemistry, Obafemi Awolowo University (OAU), Ile-Ife, Osun State, Nigeria; 3. Department of Medical Biochemistry, Obafemi Awolowo University (OAU), Osun State, Nigeria; 4. Department of Medicine, Obafemi Awolowo University (OAU), Ile-Ife, Osun State, Nigeria; 5. Department of Surgery (Ophthalmology Unit), Obafemi Awolowo University (OAU), Ile-Ife, Osun State, Nigeria.

Purpose
In recent times, Diabetes mellitus had been reported as the most prevalent disease worldwide. Its prevalence is connected to insulin resistance; a plausible link to the development of cardiovascular disease beginning at the pre-diabetes stage. Enzymatic and non-enzymatic antioxidants are recognized as potential targets for managing oxidative stress developed during diabetes and probable treatment in cardiovascular diseases in metabolic disorder patients. These enzymatic antioxidants are oxidative stress markers used in determining the glycemic index in type 2 Diabetes mellitus. The study therefore aimed at measuring and evaluating the levels of these antioxidants as well as determining the relationship between these parameters and glycemic control of Type II diabetic patients in Southwest, Nigeria.

Methods
A total of one hundred and fifteen (115) subjects were involved in this case-control study based on the formula described by Unwin et al (2010). 65 diabetic subjects were compared with 50 normal healthy controls in terms of their biochemical profiles, oxidative stress markers and glycemic index profile. Venous blood of the patients and control were collected aseptically and the plasma was collected after centrifugation at 4000 revolutions per minute (rpm) for 20 mins. Erythrocytes were separated from plasma and washed three times with 250 mM Tris HCl, pH 7.4. Glycosylated hemoglobin was determined using Clover HbA1c analyzer while fasting blood glucose was determined using glucose oxidase kit (Randox UK). Plasma aliquots were frozen at -80°C and used to analyze catalase, superoxide dismutase (SOD), protein concentration, glutathione s-transferase (GST), zinc, copper, magnesium, vitamins C and E. Total protein concentration was measured by Bradford Method. SOD activity was determined by the method of Misra and Fridovieh (1972). Catalase activity was determined by a spectrophotometrical measurement of the rate of hydrogen peroxide decomposition by the method of Aebi et al (1973). GST was assayed by monitoring the amalgamation of 1-chloro-2,4-dinitrobenzene (CDNB) and reduced glutathione (GSH) by the method described by Habig et al (1974). Plasma levels of zinc, copper and magnesium were determined using standard procedures. Vitamin C levels were measured by redox titration with 2, 6-dichlorophenolindophenol against plasma aliquots acidified by trichloroacetic acid.
while that of vitamin E were determined by a modified spectrophotometric method of Rutkowski et al (2005).

**Results**

Mean age of the diabetic patients ranged from 37 – 90 years was 61.03±11.06 years while that of the control group was 58.44±10.80 years with a range of 37–65 years. Thirteen diabetic subjects were found to have good glycemic control (HbA1c <7.0 %), twenty-eight subjects had fair glycemic control (HbA1c between 7.0–9.0 %) while the remaining twenty-four diabetic subjects had poor glycemic control (HbA1c >9.0 %). The body mass index (BMI) of diabetic patients ranged from 18.69 kg/m2 to 42.76 kg/m2 with a mean of 27.34±10.90 kg/m2. The mean values of fasting blood sugar, glycosylated hemoglobin (HbA1c), catalase, SOD, glutathione s-transferase, magnesium, zinc, copper, vitamin C and E for diabetic patients in this study are 9.12±3.86 mmol/L, 8.83±2.34 %, 48.20±2.69 units/mg protein, 53.22±2.44 units/mg protein, 1.35±0.09 units/mg protein, 0.64±0.07 mmol/L, 6.61±4.11 µmol/L, 19.07±8.96 µmol/L, 77.79±24.98 µmol/L and 1.47±1.10 µmol/L respectively while those for the healthy control subjects are 4.77±0.45 mmol/L, 63.22±1.83 units/mg protein, 62.54±2.68 units/mg protein, 3.47±0.11 units/mg protein, 0.77±0.03 mmol/L, 15.01±4.26 µmol/L, 16.89±6.25 µmol/L, 72.68±15.33 µmol/L and 4.79±1.38 µmol/L respectively without HbA1c.

Significant differences at p<0.05 and p<0.01 were found in the levels of fasting blood sugar (p = 0.286*, r = 0.021), SOD (p = -0.300**, r = 0.01), Zn (p = -0.443**, r = 0.030), Mg (p = -0.405**), catalase (p = -0.412**, r = 0.001), GST (p = -0.223*, r = 0.017) and vitamin E (p = -0.524**) when the mean values of the diabetic patients and that of the control subjects were compared. A significant difference at p<0.05 was also found when the mean values of diabetic patients without microvascular complications, with one microvascular complication and with two microvascular complications were compared and correlated. This study however did not show correlation in the antioxidant status and glycated hemoglobin.

**Conclusions**

Changes in glutathione-S-transferase enzyme activities were observed in diabetes patient. The pattern of alterations in antioxidant trace elements and vitamins of diabetic patients in this study appear to be a consequence of diabetes itself, and are not predicted by glycemic status or the presence of microvascular complications. Therefore, supplementation with zinc, magnesium and higher doses of vitamin C and vitamin E could be used by physicians in the management of Diabetes mellitus. Conclusively, additional larger scale studies are needed in precise evaluation of the role of oxidative stress markers in Type II diabetic patients.