International Journal of Healthcare Sciences ISSN 2348-5728 (Online)
Vol. 3, Issue 1, pp: (426-430), Month: April 2015 - September 2015, Available at: www.researchpublish.com

Serum levels of Vitamin E in Streptozin-Induced Diabetic Rats Treated with Watermelon Methanolic Seed Extracts

[1]Muhammad, Y., [2]Zainab, A.D., [3]Bukoye, A. O, [4]Ibrahim, M.K, [5]U. Wali, [6]Yeldu, M.H, [7]Muhammad, Y., [8]Ahmad, MB

[1, 5, 6] Department of chemical Pathology, Faculty of Medical Laboratory Science, Usmanu Danfodiyo University, Sokoto
[2, 3, 4,7Laboratory] department, Rasheed shekoni specialist hospital, Dutse, Jigawa-Nigeria
[8]Department of chemical Pathology, Bayero University, Kano

Abstract: **The discovery that oxidative stress plays a central role in the development and progression of Diabetes along with its complications is well established. Diabetes, group of disorders attributed to hyperglycaemia with abnormal lipid metabolism remains one of the leading cause of mortality throughout the world. Vitamin E is undoubtly an effective antioxidant which assist the body in scavenging free radicals thereby alleviating oxidative stress. The present study aimed to evaluate the serum levels of vitamin E in Streptozocin induced Diabetic Albino rats treated with watermelon methanolic seed extracts. In this study, twenty five albino rats weighing averagely 150g were randomly divided into five groups of five rat each (n=5). Group A= Non Diabetic control; Group B= Diabetic control; Group C, D and E = Diabetic treated with 50mg, 100mg and 200mg of watermelon seed extracts respectively. The rats were housed in a healthy condition for a period of a week to acclimatize before the commencement of the experiments, food and water ad libitum. Diabetes was induced by single intraperitonial injection of 60mglkg body weight. Vitamin E levels were determined using method Neil and Pearson (1967) after 28 day treatment. The results indicates that serum vitamin E were significantly reduced (P<0.005) in diabetic group when compared to those treated with (100mg, 200mg of extracts) and control group. In conclusion, there were significantly lower serum vitamin E levels in streptozocin induced diabetic rats, a signal that diabetics are more prone to oxidative stress.**

Keywords: **Diabetes, Vitamin E, Watermelon seed extract Free radicals, Antioxidants.**

1. INTRODUCTION

The active form of vitamin E was isolated from wheat germ oil and named tocopherol (*tokos*=child birth; *pheros*= to bear; *ol*= alcohol), initially it was discovered in an animal experiment that vitamin E deficiency leads to infertility and therefore came to be known as anti-infertility vitamin (Vasudevan, 2010). It is a family of eight naturally occurring homologous four tocopherol homologues (α β γ δ) and four tocotrienols, alpha-tocopherol is the most abundant and biologically active amongst them (Vasudevan, 2010). Diabetes mellitus is a metabolic disorder resulting from the defect in insulin secretion and action. It is not a single disease entity rather, a multisystem metabolic disorder showing common underlying feature of hyperglycemia (Maitra, *et al* 2004.). Diabetes is widely known to induce metabolic derangement leading to oxidant-antioxidant imbalance (Baynes, 1991). Hyperglycemia is associated with increase free radicals generation leading to oxidative stress; which is defined as persistent imbalance between the production of highly reactive molecular species (oxygen and nitrogen) and antioxidant defenses (Evans, *et al.,* 2003). Hyperglycaemic state increases oxidative stress in different pathways either by non enzymatic glycosylation reaction, mitochondrial electron transfer system or hexosamine pathway which is enhanced in the diabetic state (Kawahito *et al.*, 2009).

Epidemiological studies revealed that vitamins may play a protective role in the development of chronic diseases including cardiovascular diseases, diabetes, cancers and inflammatory diseases (Coyne, 2005).

Tocopherols and tocotrienols mimics process of lipid peroxidation because they mop off or scavenge lipid peroxyl radicals faster than the radical reacts with available adjacent fatty acid side chains or membrane protein.

2. MATERIALS AND METHODS

Plant materials:

The fruit pulps of watermelon were purchased from ultra modern market, Dutse, Jigawa State, Nigeria. The plant was identified and authenticated by a specialist in the Department of Biological Sciences, Faculty of Sciences. Federal University Dutse, Jigawa-Nigeria

Seed extract preparation:

The fruit pulps were broken, flesh was removed and the seeds were air dried then crushed with a pestle and mortar and later sieved to obtain the powder. The powdered sample was weighed and soaked in methanol for 48hrs, using whatman filter paper No. 1 the solution was filtered and the methanol was evaporated with rotary evaporator.

Experimental animals:

Albino rats weighing averagely 150g obtained from Department of Pharmacology, ABU Zaria were used in this study. The animals were housed in healthy condition at a constant environment with a 12-h light/dark cycle and nutritionally balanced pellets and water *ad libitum* to acclimatize for a period of two weeks before the commencement of the experiment

Induction of Diabetes:

Diabetes was induced by single intraperitonial injection of freshly prepared solution of STZ at a dose of 60mg/kg body weight in 0.1 M citrate buffer, pH 4.5. Diabetes was confirmed by measuring 10-h fasting blood glucose. Animal with blood glucose ≥ 280mgd/l were considered diabetic

Study design:

Twenty albino rats were randomly allocated into five groups of five rats each (n=5).

Group A: Non Diabetic Control rats;

Group B: Diabetic Control (Untreated) rats;

Group C: Diabetic rats treated with 50mg of watermelon seed extract.

Group D: Diabetic rats treated with 100mg of watermelon seed extract.

Group E: Diabetic rats treated with 200mg of watermelon seed extract.

The administration of extract was totally by gavage. The serum concentrations of Vitamin E of all the animals in each group were determined after the 28th treatment. On the 29th day of treatment and following an overnight fast, the animals were sacrificed under Diethyether anesthesia and blood specimens were collected directly from heart.

Biochemical analysis:

The serum vitamin E was analyzed in accordance with the method of Neil and Pearson, (1963). Alpha tocopherol is oxidized to tocopheolquinone and Fe^{2+} by $Fecl_2$. The resultant Fe^{2+} will form complex with αα-diphyridyl to produce a red color complex which is measured at 520nm after addition of ferric chloride

Chemicals used:

All reagents used are of analytical grades obtained from Sigma (St lious, USA) unless otherwise stated.

3. RESULTS

Table1: Serum levels of Vitamin E in STZ-Induced Diabetic Albino rats

GROUPS	VITAMIN E (mg/dl)
NDC	0.83±0.08
DC	0.21±0.03
Diabetic+50mg Extracts	0.23±0.04
Diabetic+100mg Extracts	0.53±0.07
Diabetic+200mg Extracts	0.73±0.06

Results are presented as Mean±SD; n= Number of rats; NDC= Non Diabetic control; Diabetic control.

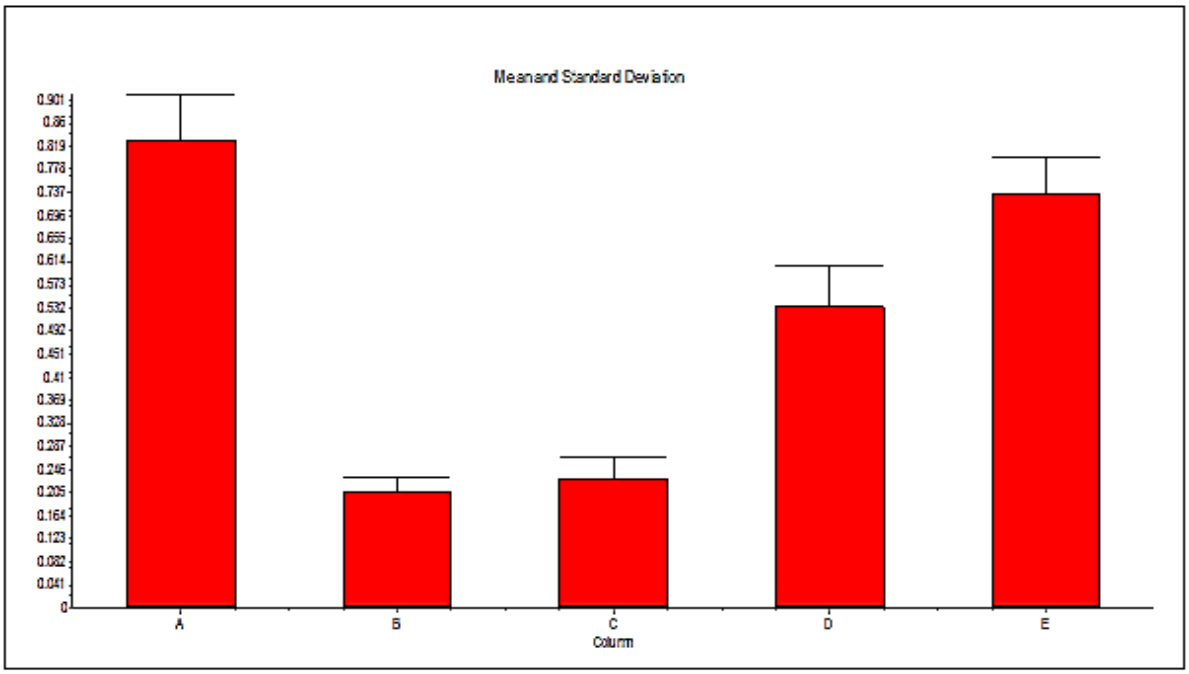

Figure 1: Distribution of Serum levels of Vitamin E in Non Diabetic and Diabetic rats treated with 50mg, 100mg and 200mg of Watermelon Seed Extracts.

A= Non Diabetic control; B= Diabetic control (untreated); C=Diabetic+50mg; D=Diabetic+100mg; E= Diabetic+200mg.

4. DISCUSSION

Antioxidants have been in use for the treatment and management of diabetes and its complications. Many studies have shown clearly that serum levels of antioxidant vitamin E is reduced in diabetes (Peerapatdit *et al*., 2006; Murugan *et al*., 2006; Lee *et al*., 2007; Ramesh et al., 2006; Wu *et al*., 2007). When exercise is given to rats with STZ-induced diabetes in addition to Vitamins C and E, it was observed that lipid peroxidation was significantly reduced, glutathione peroxidase (GSH-Px) was increased and reduced glutathione (GSH) level was decreased (Kutlu *et al*., 2005).

The results of the current study, shows a significant decreased of serum vitamin E ($P<0.005$) in diabetic compared to control group. This is because of increased oxidative stress in diabetes that are associated with excessive production of free radicals leading to consequent consumption of vitamin E in order to attenuate the effect of radicalized molecules that precipitated the diabetic complications.

Studies suggest that vitamins (such as C and E) are important in preventing or alleviating the complications of diabetes mellitus (Harding *et al*., 2008). Vitamin C and E not only reduce the risk of thromboembolism in patients with diabetes-related hypertension Haidara *et al*., (2004) but also concert a positive effects on wound healing (Musalmah *et al*., 2005).

Vitamin E is a major fat soluble antioxidant that acts as a scavenger for free radicals by inhibiting the chain reactions of lipid peroxidation. It works by donating the hydrogen from its hydroxyl group on its ring structure to free radicals making them uncreative (Punithavatki, *et al*., 2008). Vitamins C and E have been shown to prevent the teratogenic effects in diabetic rats and autoimmunity of cells in babies (Uusitalo *et al*., 2008; Cederberq *et al*., 2005). Vitamins C and E can be used as antioxidants separately or in combination. Both vitamins act synergistically (Naziroglu *et al.*, 2004; Kutlu *et al.*, 2005)

Administration of 50mg of watermelon seed extracts to diabetic rats revealed no significant difference in the serum levels of vitamin E when compared to diabetic control, this is because the concentration provides little or no impacts on the antioxidant status, however diabetic rats treated with 100mg and 200mg of seed extracts shows a statistical increase in the serum vitamin E.

A positive relation has been established between high serum vitamin E level and reduction in diabetic complications (Harding et al., 2008), vitamin E is very effective in glycemic control, lowering the HbA1c levels (Ihara et al., 2000) and critical in preventing the hypertropic effects of hyperglycemia (Nascimento et al., 2005).

5. CONCLUSION

There were significantly high serum levels of Vitamin E in diabetic rats treated with 100mg and 200mg of watermelon seed extracts compared to diabetic untreated.

It is therefore recommended that diabetic patient should take more of wheat germ oil, sunflower oil, safflower oil, fish liver oil and fruits which are natural sources of vitamin E with the view to mitigating the possible oxidative stress thereby increasing life span

REFERENCES

[1] Baynes, JW (1991). Role of oxidative stress in development of complications in diabetes. *Diabetes*; 40:405-412

[2] Cederberq J, Erikssan UJ (2005). Antioxidative treatment of pregnant diabetic rats diminishes embryonic dysmorphogenesis. Birth Defects *Res. A. Clin. Mol. Teratol*. 73: 498-505

[3] Coyne, T. (2005): Antioxidant, *Am. J. Clin. Nutr* 82: 3685 – 3698.

[4] Evans, JL., Goldfine, ID., Maddux BA., Grodsky, GM (2003). Are oxidative stress activated signaling pathways mediation of insulin resistance and beta cell dysfunction? *Diabetes* 2003; 52: 1-8

[5] Haidara MA, Khloussy H, Ammar H, Aal Kasem LA (2004). Impact of alpha-tocopherol and vitamin C on endothelial markers in rats with streptozotocin-induced diabetes. *Med. Sci. Monit*. 10: BR41-6.

[6] Harding AH, Wareham NJ, Bingham SA, Khaw K, Luben R, Welch A, Forouhi NG (2008). Plasma vitamin C level, fruit and vegetable consumption and the risk of new-onset type 2 diabetes mellitus: The European prospective investigation of Cancer – Norfolk prospective study. *Arch. Inter. Med*. 168: 1485-1486

[7] Ihara Y, Yamada Y, Toyokuni S, Miyowaki K, Ban N, Adachi T, Kurae A, Iwakuru T, Kubota A, Hiai H, Seino Y (2000). Antioxidant alphatocopherol ameliorates glycemic control of GK rats, a model of type 2 diabetes. *FEBS Lett*. 473: 24-6.

[8] Kawahito, S, Kitahata, H, Osita, S (2009). Problems associated with glucose toxicity: role of hyperglycaemia-induced oxidative stress. *World journal of gastroenterology* 15(33) 4137-4142

[9] Kutlu M, Nazıoglu M, Simsek H, Yılmaz T, Kukner AH (2005). Moderate exercise combined with dietary vitamins C and E counteracts oxidative stress in the kidney and lens of streptozotocin-induced diabetic rat. *Int. J. Vitam. Nutr. Res*. 75: 71-80.

[10] Lee EY, Lee MY, Honq SW, Chunq CH, Honq SY (2007). Blockade of oxidative stress by vitamin C ameliorates albuminuria and renal sclerosis in experimental diabetic rats. *Yonsei Med. J.* 48: 847-55.

[11] Naziroglu M, Simsek M, Simsek H, Aydilek N, Özcan Z, Atılgan R (2004). The effects of hormone replacement therapy combined with vitamins C and E on antioxidants levels and lipid profiles in postmenopausal women with type 2 diabetes. *Clin. Chem. Acta*. 344: 63-71.

[12] Nield, J.B. and Pearson, W. N. (1967): Macro and Micro method of determination of serum vitamin E. Using Triflouro acetic acid. *Nutr* 79: 10-20.

[13] Maitra A, Abbas, Robbins and Cotran (2004). Pathologic basis disease, 7th ed.. P 1191-1192).

[14] Murugan P, Pari L (2006). Antioxidant effect of tetrahydrocurcımin in streptozotocin nicotinamide induced diabetic rats. *Life Sci.* 79: 1720- 1728

[15] Musalmah M, Nizrana MY, Fairuz AH, Noor Aini AH, Azian AL, Gapor MT, Wan Ngah WZ (2005). Comparative effects of paim vitamin E and alpha-tocopherol on healing and wound tissue antioxidant enzyme levels in diabetic rats. *Lipids* 40: 575-80.

[16] Nascimento Gomes G, Barbosa FT, Radaeli RF, Cavaral MF, Mello Aires M, Zaladek Gil F (2005). Effect of D-alpha-tocopherol on tubular nephron acidification by rats with induced diabetes mellitus. *Braz. J. Med. Biol. Res*. 38: 1043-51.

[17] Peerapatdit T, Patchanans N, Likidlilid A, Poldee S, Sriratanasathavorn C (2006). Plasma lipid peroxidation and antioxidant nutrients in type 2 diabetic patients. *J. Med. Assos. Thai. 89 Suppl*. 5: 5147-55.

[18] Punithavatki VR, Anuthama R, Prince PS (2008). Combined treatment with naringin and vitamin C ameliorates streptozotocin-induced diabetes in male Wistar rats. *J. Appl. Toxicol*. 28: 806-813

[19] Ramesh B, Saravanan R, Pugalendi KV (2006). Effect of dietary substitution of Groundnut oil on blood glucose, lipid profile, and redox status in streptozotocin-diabetic rats.

[20] Yale J. Biol. Med. 79: 9-17. Ritz E (2006). Diabetic nephropathy. *Saudi J. Kidney Dis. Transpl*. 17: 481-490

[21] Uusitalo L, Nevalainen J, Niinistü S, Alfthan G, Sandvall J (2008). Serum alpha and gamma-tocopherol concentrations and risk of advanced beta cell autoimmunity in children with HLA-conferred susceptibility to type 1 diabetes mellitus. *Diabetologia* 51: 773-80.

[22] Vasudevan, DM and SreeKumari,S. (2010). Textbook of Biochemistry for Medical Students. "Fat soluble vitamins." 6th edition.Jaypee brothers Medical publishers, New Delhi. Pp 291

[23] Wu JH, Ward NC, Indrawan AP, Almeida CA, Hodgson JM, Porudfoot JM, Puddey IB, Croft KD (2007). Effect of _-tocopherol and mixed tocopherol supplementation on markers of oxidative stress and inflammation in type 2 diabetes. *Clin. Chem*. 53: 511-519.