EFFECTS OF PALM OIL ON SOME OXIDATIVE INDICES OF ALLOXAN INDUCED DIABETIC RABBITS

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ABSTRACT

The effects of palm oil on oxidative indices of alloxan induced diabetic rabbits were investigated. The result obtained showed that palm oil significantly decreased (P < 0.05), lipid peroxidation in diabetic treated animals. The vitamin C (antioxidant vitamin) level increased significantly (P < 0.05) in the supplemented group but decreased significantly (P < 0.05) in non supplemented group. The glucose levels in both the diabetic supplemented group and non supplemented group were not significantly different (P > 0.05). The results indicate that palm oil supplementation decreased the level of lipid peroxidation but increased the level of vitamin C, an indication that palm oil can attenuate oxidative stress generated in diabetic condition. This result may suggest that supplementation of palm oil may be effective in the management of diabetes mellitus.

Keywords: Red palm oil, Diabetes mellitus, Antioxidant, Oxidative stress, Lipid peroxidation

INTRODUCTION

The pathogenesis of many diseases involve free radical – mediated lipid peroxidation of biological membrane (Eze, *et al*., 1993; Eze, 1992; Ogugua, 2000) Diabetes mellitus and its complications have been associated with oxidative stress (Okamoto, 1981; Ogugua, 2000). It is reported that the oxidative destruction of the islets of Langerham of the pancreas by alloxan leads to diabetes mellitus and is free radical-mediated (Ofordile, 1987; Aruoma, *et al.,* 1991; Ogugua, 2000; Galluzzo, *et al.,* 1990). Membranes are prone to oxidation by reactive oxygen species (ROS) and because of the devastating complications and health hazard associated with diabetes mellitus, its management has continued to occupy researches in both medicine and related discipline (Eze, 1992; Ogugua, 2000; Aruoma, *et al.,* 1991)

Diabetes mellitus can only be managed or prevented but not cured. The role of antioxidant nutrients for the management of various diseases associated with oxidative stress has been well documented (Halliwell, *et al.,* 1992, Gutheridge, 1994.) Vitamins A, C, E and B carotene have been found useful (Muma, 1994; Ogugua, 1994; Gutheridge, 1994). Thus adequate dietary intake of vitamin E, a major lipid soluble inhibitor of peroxidation may be important in inhibiting the development of disease conditions including diabetes mellitus. Palm oil has been found to contain a lot of vitamin E and other lipid soluble vitamin nutrients (Atroshi, *et al.,* 1992; Choo, *et al.,* 1992; Packer, 1992; Gutheridge, 1994).

Since this vegetable oil is very common, affordable and used by majority of people across the globe especially in the tropics, its use as antidote to prevent some oxidative stress related diseases and complications is advocated. As reported else where (Choo, *et al*., 1992; Atroshi, *et al*., 1992) that palm oil contains mainly vitamin E, a major chain-breaking antioxidant in the membrane, it is the thrust of this work to study the effects of palm oil supplementation on some oxidative indices in alloxan induced diabetic rabbits. The outcome of the work may become useful in the management of human diabetics.

MATERIALS AND METHODS

Palm oil was bought from Nsukka local market and used for the experiment. Twelve albino rabbits weighing (2.5 kg on the average) were bought from Chigbo rabbitary Research Centre Awka Anambra State, Nigeria. The animals were kept for two weeks in laboratory to acclimatize with the environment. They were grouped into three groups of four rabbits each namely group 1 (normal rabbits), group 2 (diabetic rabbits but not treated with palm oil) and group 3 (diabetic rabbits but treated with palm oil). Diabetes was induced by administration of alloxan at 180 mg/kg body weight. The test diabetic animals (group 3) were given 5 ml of palm oil orally twice a day for two weeks. Blood samples were collected from the ear veins of the animals for the assay.

Malondialdehyde (MDA) level was estimated by the method of Albro *et al* (1986) and Das *et al* (1990). The thiobarbituric acid procedure employs the reaction of thiobarbituric acid with malondialdehyde to form a red chromogen which absorbs at 532nm. The concentration is

proportional to the level of peroxidation. Vitamin C level was determined by the method of Tietz (1970). This involves the oxidation of ascorbic acid (vitamin C) in the presence of combined colour reagent. The hydrozone resulting from this procedure dissolves in strong sulfuric acid solution to produce a red complex which absorbs at 500 nm. Glucose level was estimated according to 0-Toluidine method of Cooper & McDaniel (1970) in which the glucose in the sample when heated with O-Toluidine regent gives a blue-green colouration, the intensity of which is proportional to glucose concentration.

The results were analysed using analysis of variance (ANOVA) and expressed as mean ± SD.

RESULTS AND DISCUSSION

Table 1 shows that glucose level increased in diabetic condition (DNT group 2) compared with both the normal rabbits (group 1) and rabbits supplemented with palm oil. It was observed that palm oil slightly decreased glucose level in group 3 (DT).

That glucose levels increase in diabetic condition has been variously reported (Hamme, *et al.,* 1991; Takuncu, *et al.,* 1998; Sharpe, et al., 1998). Also, increase in glucose level has recently been associated with oxidative stress (Atkinson and Maclaren, 1990; Yadar, *et al.,* 1997; Ogugua 2000). Thus, the increased glucose level in group 2 (diabetic not treated group) is a result of intrinsic oxidative stress in diabetic condition. The slight reduction in the glucose level which was not significantly different ($P > 0.05$) is an indication that palm oil contains antioxidant which possibly countered oxidative stress in the animals. Earlier reports (Atroshi, *et al*., 1992; Choo, *et al*., 1992) show that palm oil contains other antioxidant vitamins – A and B carotene besides high level of vitamin E and, these might have acted synergistically, to reduce blood glucose. This observation lay credence to the report of Chung *et al* (1992) that these nutrients played protective roles against oxidative stress in alloxan induced diabetic rats. The very slight increase in glucose level in the normal group as experiment progressed could be due to increased oxidative stress resulting possibly from repetitive bleeding. These inferences corroborate a stipulation by Rey and Besedovsky (1989) that repetitive bleeding increases oxidative stress and glucose level.

Malondialdehyde (MDA) level increased in diabetic rabbits (group 2) compared with the normal. This increase is significant ($P < 0.05$). Diabetic condition has been linked with oxidative stress (Elhadd, *et al.,* 1999), and increased lipid peroxidation product (Ogugua, 2000), expressed as malondiadehyde level (MDA). Thus, the high level of MDA in group 2 – diabetic not treated rabbits – could be explained on the basis of oxidative stress mediated lipid peroxidation while the reduction in the level in group 3 (diabetic supplemented group) could be a result of the antioxidant property of palm oil. This property is conferred to palm oil by its possession of high level of a-tocopherol and other antioxidant vitamin (Packer, 1992; Atroshi, *et al.,* 1992).

Table 1: Effect of red palm oil on blood glucose, lipid peroxidation and vitamin C levels during the experiment

Group	Glucose level mmol/L
1	8.10 ± 0.30 6.40 ± 0.40[a] ,7.66 ± 0.46[b]
2	13.68 ± 0.48* 6.48 ± 0.28[a], 12.50 ± 0.35[b]
3	12.56 ± 0.36* 6.65 ± 0.52[a], 14.20 ± 0.60[b]
	Lipid peroxidation level mmol/ml plasma
1	7.90 ± 0.22
2	13.73 ± 0.41**
3	9.95 ± 0.20**
	Vitamin C level mg/100m
1	0.83 ± 0.18
2	0.62 ± 0.11**
3	0.69 ± 0.13**

*Group 1 is the normal rabbits (non diabetic), Group 2 is diabetic rabbits not supplemented with palm oil, Group 3 is the diabetic rabbits supplemented with palm oil, [a] indicates basal blood glucose level [b] indicates blood glucose before treatment, *p> 0.05, **p<0.05, Rabbits with blood glucose 10mmol/l and above were considered diabetic*

Vitamin E supplementation has been reported to be efficacious in reducing oxidation in diabetics (Bethesda, 1991). It is evidenced in the present study that this antioxidant nutrient in palm oil scavenged free radicals and hence reduced oxidative stress in diabetic treated rabbits. This corroborate a report that palmvitee capsule – a vitamin E capsule from palm oil – reduced serum cholesterol level in oxidative condition (Jacobson, *et al.,* 1990).

The low vitamin C level in group 2 is expected as oxidative stress operates in diabetic condition (Ogugua, 2000). Vitamin C is always the first antioxidant nutrients that counters oxidation at cytosol level and are thus depleted (Frei, 1991, Das and Thurhnam, 1992). The increase in vitamin C level in group 3 again suggests that palm oil contains some antioxidants that helped to scavenge free radicals in the system. This action probably spared the available vitamin C in the

system. It could be that vitamin E being helped by other antioxidant components in the palm oil prevented the generation of free radicals and subsequent oxidation. Similar synergistic action of antioxidant nutrient has been reported (Chiu, *et al.,* 1982).

The work in essence shows that palm oil contains some nutrients possibly antioxidants capable of suppressing oxidative stress. Research elsewhere suggests that vitamin E is a major ingredient. The work thus suggests that palm oil may be a good antioxidative nutrient as its supplementation decreased lipid peroxidation but increased vitamin C level. The effect on glucose level was not pronounced, suggesting that the effect could be more on the complications of diabetes.

In conclusion palm oil may help to attenuate diabetes mellitus and possibly diabetes complications. Its use may then be suggested in the management of diabetic condition and in diseases associated with oxidative stress.

REFERENCES

ALBRO, P. W. CORBETT, J. T. and SCHROEDER, J. L. (1986). Application of the thiobarturic assay to the measurement of lipid peroxidation products in microsomes. *Journal of Biochemistry*. *261:* 4889 – 4894.

ARUOMA, O. I., KAUR, H. and HALLIWELL, B. (1991). Oxygen free radicals and human disease. *Journal of Royal Society Health, 8:* 172 – 177.

ATKINSON, M. A. and MODOREN, N. K. (1990). What causes Diabetes? *Scientific American, July*: 42 – 47.

ATROSHI, F., ANTILA, E., SANKARI, S., TREUTHARAT, J., GAPOR, A., SALONIEMI, H. and WEITER M. T. (1992). Palm oil vitamin effects in hypercholesterolemia. Pages 243 – 254. *In:* ONG, A. S. H. and PACKER, L. (Eds.). *Lipid soluble antioxidants: biochemistry clinical application.* Birkhauser Verg, Boston.

BETHESDA, M. D. (1991). Guidelines for the diagnosis and management of Asthma. *National Institute of Health, US Department of Health and Human Services Publications, 91:* 30 – 31.

CHOO, Y. M., YAP, S. C., OOI, C. K., ONG, A. S. H. and GOLE, S. H. (1992). Production of palm oil carotenoid concentrate and its potential application in nutrition. Pages 243 – 254. *In:* ONG, A. S. H. and PACKER, L. (Eds.). *Lipid soluble antioxidants: biochemistry and clinical applications*. Birkhauser Verg Boston.

CHIU, D., VICHINSKY, E. Y. M., KLEMAN, K. and LUBIN, B. (1982). Peroxidation of vitamin E and sickle cell anaemia. *Annals of New York Academy of Sciences, 239:* 323 – 335.

CHUNG, Y. M. I., CHIU, P. M. M. and WONG, H. L. (1992). Modification of alloxan diabetes in rats by vitamin A status. Pages 265 – 273. *In:* ONG, A. S. H. and PACKER, L. (Eds.). *Lipid soluble antioxidants: biochemistry clinical application.* Birkhauser Verg, Boston.

COOPER, G.R. and McDANIEL, V. (1970). Assay methods. Pages 159 – 170. *In:* McDONALD, R. P. (Ed.). *Standard methods for clinical chemistry*. John Wiley and Sons, New York.

DAS, B. S. THURNHAM, D. I. PATNACK, J. K. DAS, D. B. SATPATHY, R., and BASE, T. K. (1990). Increased plasma lipid peroxidation in riboflavin deficient malaria – infected children. *American Journal Clinical Nutrition*, *51:* 859 – 863.

DAS, B. S. and THURNHAM, D. I. (1992). Plasma lipid peroxidation in *Plasmodium falciparium* malaria. Pages 397 – 405. *In:* ONG, A. S. H. and PACKER, L. (Eds.). *Lipid soluble antioxidants: biochemistry clinical application.* Birkhauser Verg, Boston.

ELHADD, T. A., KHAN, F., KIRK, G., MCLAREN, M. and NEWTON, R. W. (1999). Influence of poverty on endothelial dysfunction and oxidative stress in young patients with type 1 diabetes. *Diabetes Care, 21:* 1990 – 1996.

EZE, M. O. (1991). Production of superoxide by macrophages from Plasmodium chandandi infected mice. *Cytobios, 66:* 93 – 104.

EZE, M. O. (1992). Membrane fluidity, ROS and Cell mediated immunity; implications in nutrition and disease. *Medical Hypotheses, 37:* 220 – 224.

FREI, B. (1991). Ascorbic acid protects lipid in human plasma and low density lipoprotein against oxidative damage *American Journal Clinical Nutrition, 54:* 1113 – 1118.

GALLUZZO, A. GIORDANO, C. and BOMPIANA, G. D. (1990). Cell-mediated immunity in diabetic pregnancy. Pages 273 – 278. *In: Immunology of normal and diabetic pregnancy.* John Wiley and Sons New York.

GUTHERIDGE, J. M. C. (1994). Reactivity of hydroxyl and hydroxyl – like radical discriminated by release of TBARS from deoxysitrose, dudeosides and Beuroate. *Biochemistry, 224:* 761 – 767.

HAMME, H. R. MARTINS, S. FEDERLIN, K., GEISEN, K. and BROWNEE, M. (1991). Aminoguanidine treatment inhibits the development of experimental diabetes retinopathy. *Proceedings of National Academy of Sciences, 88:* 11555 – 11558.

JACOBSON, J. M., MICHAEL, J. R., JAFRI M. H. and CUISTNER, G. H. (1990). Antioxidants and antioxidant enzymes protects against pulmonary oxygen toxicity in the rabbit. *Journal Applied Physiology, 68:* 1252 – 1259.

MUMA, A. F. (1994). *Lipid peroxidation in malaria patients.* MSc Dissertations, Department of Biochemistry University of Nigeria, Nsukka. 145 pp.

OFORDILE, P. M. (1987). *Possible interaction of experimental diabetes with environmental toxins, biochemical and histopattiological evaluations*. MSc Dissertation, University of Nigeria, Nsukka. 108 pp.

OGUGUA, V. N. (1994). *Lipid peroxidation and antioxidant status in sicklers carriers and normal individuals.* MSc Dissertation, Department of Biochemistry, University of Nigeria, Nsukka. 128 pp.

OGUGUA, V. N. (2000). *Assessment of some parameters of oxidative stress in alloxan induced diabetic rabbits.* PhD Thesis, Department of Biochemistry, University of Nigeria, Nsukka. 205 pp.

OKAMOTO, H. (1981). Molecular basis of experimental diabetes degeneration, oncogenesis and regeneration of pancreatic Islets of Langerhams. *Bioassay, 2:* 15 – 21.

PACKER, L. (1992). New horizons in vitamin E research – the vitamin E cycle, biochemistry, and clinical applications. Pages 1 – 16. *In:* ONG, A. S. H. and PACKER, L. (Eds.). *Lipid soluble antioxidants: biochemistry and clinical applications*. Birkhauser Verg, Boston.

REY, A. D. and BESEDORSKY, H. (1989). Antidiabetic effects of interlukin. *Proceedings of National Academy of Sciences, 86:* 5943 – 5947.

SHARPE, P. C., YUE, K. K. M., CATTERWOOD, M. A., McMARTER, D. and TRIBBLE, E. R. (1998). The effects of glucose induced oxidative stress on growth and extracellular matrix gene expression of vascular smooth muscle cells. *Diabetologia, 41:* 1210 – 1219.

TAKUNCU, N. B., BCYRAKTAR, M. and VARLI, K. (1998). Reversal of defective nerve conductions with vitamin E supplementation in type II diabetes. *Diabetes Care, 21:* 1915 – 1918.

TIETZ, N. N. (1970). Carbohydrate. Pages 174 – 176. *In:* Sauders, W. B. (Ed). *Fundamental of Clinical Chemistry*. W. B. Sauders Company, London.

YADAR, P., SARKAR, S. and BHATNAGARD, D. (1997). Action of *Capparis deciduas* against alloxan induced oxidative stress and diabetes in rat tissue. *Pharmacology Research, 36(3):* 221 – 228.