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Research Article

Alteration in Plasma Tocopherol Levels in the Patient with Different Stages of Essential Hypertension

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Abstract: Free radicals and subsequent damage in the form of lipid peroxidation have been implicated in the pathogenesis of variety of cardiovascular diseases including essential hypertension. However, its precise etiology is unknown. These free radicals are effectively controlled by antioxidants but under pathophysiological condition these antioxidant level changes which enhance further deterioration. In this context, a study was done to assess the levels of plasma tocopherol, a potent non-enzymic antioxidant and malondialdehyde (marker of lipid peroxidation) in Hypertensive subjects and to determine the variation in their levels with severity of disease. In the present study, plasma tocopherol level and Malondialdehyde levels were measured in 60 hypertensive subjects (30-55 years) which were categorized into three groups as prehypertension, Stage I HT and Stage II HT, depending upon their blood pressure and statistically compared it with that of 20 healthy individual, served as control. Plasma tocopherol level was found to be significantly low in each patient group as compared to control ($p<0.001$) and inversely related to the malondialdehyde levels ($p<0.05$) and blood pressure. These findings suggest that excess production of free radicals occur in the body with subsequent rise in blood pressure as characterized by increased production of malondialdehyde and plasma tocopherol is consumed in scavenging free radical to reduce oxidative stress mediated destructions. Therefore, consumption of diet rich in vitamin E should be increased with severity of disease.
Keywords: Plasma tocopherol, Malondialdehyde, Free radicals, Oxidative stress

INTRODUCTION

Hypertension, a multifactorial complex disorder, is generally defined as persistent increase of systemic arterial blood pressure i.e. more than 120/80 mm Hg with essentially unknown etiology. Amongst various risk factors and biochemical events associated with the etiopathogenesis of essential hypertension (HT), increased production of oxygen free radicals have been identified as an important inducer of vascular disorder leading to cardiovascular complications including HT [1]. Lipid peroxidation is a free radical mediated process in which the polyunsaturated fatty acids contained in the LDL or present in the cell membrane are degraded to variety of aldehydes including malondialdehyde (MDA), a marker of lipid peroxidation [2]. These aldehydes play a crucial role in endothelial dysfunction and inhibit NO synthase activity leading to the development of HT.

Antioxidants reduce or eliminate these free radicals. Among non enzymic antioxidant, vitamin E is the most potent lipid soluble and chain breaking antioxidant which has been found to be effective in the prevention of free radical mediated biomolecular deterioration and thereby proove to be cardioprotective [3]. Its level is greatly influenced with increase in blood pressure. Kumar and Das also observed that free radicals are produced in excess amount in HT patients and altered levels of antioxidant vitamins are significantly associated with both systolic and diastolic blood pressure [4]. Although limited information is available on MDA level along with plasma tocopherol level in patients with essential hypertension, alteration in their levels with severity of disease is still obscure. Therefore, the objectives of present study were to estimate erythrocyte MDA and plasma tocopherol levels in patients with essential hypertension and to determine the variation in these levels with increase in blood pressure.

MATERIALS & METHODS

In the present study, 60 subjects of essential hypertension of either sex were taken in a patient group and 20 age matched healthy individuals were taken in control group (30-55 years). The patients were classified into 3 groups (20 patients in each group), according to "Seventh Report of Joint National Committee on High Blood Pressure". These include prehypertension [Systolic Blood Pressure (SBP) 120-139 mm Hg and Diastolic Blood Pressure (DBP) 80-89 mm Hg], Stage I HT (SBP 140-159 & DBP 90-99 mm Hg) and Stage II HT (S.B.P. ≥ 160 mm Hg & D.B.P. ≥ 100 mm Hg). A general information or pre-experimental questionnaire regarding demographic information, family history and limited physical examination including blood pressure measurement was completed from all the subjects after taking their informed consent and approval of protocol by ethics committee of college. In addition, hypertensive patients with other systemic diseases viz. diabetes mellitus, proteinurea, any renal disease and having history or evidence of antihypertensive medication and antioxidant supplements were excluded.

Fasting blood samples were collected in EDTA vials from the anticubital vein of the study group subjects and processed immediately. Erythrocyte malondialdehyde (MDA) levels were measured as thiobarbituric acid reactive substances (Sinnhuber et al. 1958), after preparation of hemolysate [5]. The heat induced reaction of MDA with thio barbituric acid (TBA) in the acid solution forms a trimethine coloured substance, measured spectrophotometrically at 532 nm.

Plasma tocopherol levels were estimated by Hashim and Schuttringer method [6]. Protein in the plasma or serum is precipitated by an equal volume of absolute ethanol; the whole mixture is subjected to extraction by an equal volume of n-heptane. α,α^1-dipyridyl is added followed by Ferric chloride reagent to the system which produces light pinkish orange color and measured it spectrophotometrically at 510 nm.

Statistical analysis

Values were expressed as Mean ± SD. The significance of mean difference between groups was compared by using Student's t test and distribution of probability (p).

RESULTS

The mean plasma tocopherol level and malondialdehyde level (MDA) of the patient and control group are depicted in Table.1.0. The mean plasma tocopherol levels in hypertensive subjects were significantly low ($p<0.001$) as compared to controls i.e. 36.5%, 54.01% & 67.2% in prehypertension, stage I HT and stage II HT patients respectively. On the other hand, erythrocyte malondialdehyde levels were found to be significantly high ($p<0.001$) 34.75%, 47.8% & 68.7% in prehypertension, stage I HT and stage II HT patients as compared to healthy controls. In addition, erythrocyte MDA levels were inversely related with plasma tocopherol levels (Table 2). These variations were increased continuously with increase in blood pressure.

Table 1: Plasma Tocopherol and Erythrocyte malondialdehyde levels in patients and control group (Mean± SD)

Sl. No.	Particulars	Control Group	Patient Group		
			Prehyper--tension	Stage I HT	Stage II HT
1.	Sample size	20	20	20	20
2.	Age (years)	30-55	30-55	30-55	30-55
3.	Systolic blood pressure (mm Hg)	116 ± 3.04	128 ± 3.20	146 ± 3.58	164 ± 3.80
4.	Diastolic blood pressure (mm Hg)	76.8 ± 2.13	84.2 ± 2.38	95.6 ± 2.52	108.0 ± 2.70
5.	Tocopherol level (mg %)	1.37 ± 0.51	0.87 ± 0.26*	0.63 ± 0.21**	0.45±0.18**
6.	Malondialdehyde level (µmol MDA/ml)	1.57 ± 0.10	2.10 ± 0.12*	2.32 ± 0.15*	2.65±0.19**

* $p < 0.05$: Significant, ** $p < 0.001$: Significant

Table 2: Correlation coefficient (r) between erythrocyte MDA level and plasma tocopherol levels in Essential hypertension subjects

Particulars	Tocopherol in Prehypertension	Tocopherol in Stage I HT	Tocopherol in Stage II HT
Erythrocyte MDA	- 0.503	-0.376	-0.295

DISCUSSION

Oxidative stress is increased in patients with essential hypertension owing to increase in free radical production. Involvement of free radicals in membrane damage via lipid peroxidation and its resultant products such as lipid radicals (L◦), lipid peroxides (LOO◦), lipid hydroperoxides (LOOH) and highly reactive aldehydes plays a crucial role in the development and progression of disease process [1-3]. In the present study, a highly significant increased levels of malondialdehyde (i.e. marker of lipid peroxidation) were observed in each patient group ($p<0.001$) as compared to healthy control which rises continuously with increase in blood pressure and clarify the etiopathogenic role of free radicals via lipid peroxidantion in HT patients. Our findings were in concordance with the findings of Tandon *et al.* [7]. According to them, lipid peroxides are toxic to the cellular components and lipid peroxidation may be responsible for vascular disorder in HT. Vasdev et.al also reported that excess endogenous aldehyde plays a major role in HT by binding sulphydryl groups of membrane proteins, altering Ca^{2+} channels and increasing cytosolic free Ca^{2+} that cause further extensive membrane damage due to the action of phospholipases and proteases, the activation of contractile proteins and the accumulation of mitochondrial calcium resulting in a vicious cycle of damage extension, peripheral vascular resistance and hypertension [8].

The production of free radicals and its culprit effect on biomolecules is effectively regulated by antioxidant reserves of body. Among nonenzymic antioxidants, vitamin E, a universal lipophilic, chain breaking antioxidant and a stabilizer of biological membranes, prevent accumulation of free radicals and decreases lipid peroxidation [9, 12]. Prithviraj and Misra have reported that α-tocopherol not retards only LDL oxidation but also inhibits smooth muscle proliferation, platelets adhesion and aggregation, expression and function of adhesion molecules, decreases the synthesis of leukotrienes and potentiates the release of prostacyclin through upregulation of cytosolic phospholipase A_2 and cycloxygenase [10].

In the present study, plasma tocopherol levels were significantly low ($p<0.05$, $p<0.001$, $p<0.001$) in each patient group as compared to controls. However they did not differ significantly with each other. Decreased levels of vitamin E could not be only due to its free radical scavenging action but also in maintaining the body antioxidant reserve and in normalization of vascular superoxide formation which prevent endothelial dysfunction. Wen *et al.* also observed reduction in vitamin C & E level with increased levels of lipid peroxides in hypertensive subjects and concluded it as a contributory event in the development of CVD in HT patients [11]. In addition, Newaz *et al.* also observed that dietary supplementation of α-tocopherol in spontaneously hypertensive rats prevents increased blood pressure; lowers lipid peroxides in plasma and enhances total antioxidant status [12].

CONCLUSION

On the basis of present study and consistent findings of previous studies we conclude that lipid peroxidation plays an etiopathological role in the development of HT and plasma tocopherol level is inversely related to increase in blood pressure. Thus, both vitamin E and malondialdehyde levels may be an excellent marker of oxidative stress in essential hypertension. As the blood pressure rises, plasma vitamin E level decreases continuously not only due to its free radical scavenging action but also in maintaining body's antioxidant reserve and in limiting the lipid peroxidation. Therefore, our study suggests that the diet with high vitamin E contents is essential for patients suffering with essential hypertension and consumption of fruit, vegetables and grains should be increased with increase in blood pressure.

REFERENCES

1. Gupta V, Saxena R, Bhattacharya I, Thakur RK, Mishra M; Lipid peroxidation mediated electrolyte imbalance and altered antioxidant status in Hypertensive smokers. Saudi Germ Hosp Med J., 2011; 5(1): 7-17.
2. Bhattacharya I, Saxena R, Gupta V; Efficacy of vitamin E in knee osteoarthritis management of North Indian Geriatric population. Therap Adv Musculo Dis., 2012; 4(1):11-19.
3. Bhattacharya I, Saxena R, Saxena R, Lal AM; Vitamin E supplementation and the markers of oxidative stress in Indian Acute Myocardial Infarction patients. Asian J Medical Sciences, 2014; 5(2): 46-53.
4. Kumar K, Das UN; Are free radicals involved in the pathobiolgy of human essential hypertension? Free Rad Res Commun., 1993; 19(1): 59-66.
5. Sinnhuber RO, Yu TC, Yu TC; Characterization of the red pigment formed in the thiobarbituric acid determination of oxidative rancidity. Food Res., 1958; 23(6): 626-630.
6. Hashim SA, Schuttringer GR; Rapid determination of Tocopherol in Macro and Micro quantities of plasma. Am J Clin Nutr, 1996; 19: 137 – 144.
7. Tandon R, Sinha MK, Garg H, Khanna R, Khanna HD; Oxidative stress in patients with essential hypertension. Nat Med J India, 2005; 18(6): 297 – 299.
8. Vasdev S, Gill V, Parai S, Longerich L, Gadag V; Dietary vitamin E supplementation lowers blood pressure in spontaneously hypertensive rats. Mol Cell Biochem, 2002; 238(1-2): 111 – 117.
9. Bhattacharya I, Saxena R, Saxena R, Lal AM; Vitamin E supplementation and markers of oxidative stress in indian acute myocardial

infarction patients. Asian Journal of Medical Science, 2014; 5(2): 46-53.
10. Prithviraj T, Misra KP; Reversal of Atherosclerosis – Fact or fiction? Cardiology Today, 2000; 6(2): 97-100.
11. Wen Y, Killaleas Mc, Gettigan P, Feely J; Lipid peroxidation and antioxidant vitamin C & E in Hypertensive patients. Irish Med Sci., 1996; 165(3): 210-212.
12. Newaz MA, Nowal NNA; Effect of α–tocopherol on lipid peroxidation and total antioxidant status in SHR. Am J Hypert., 1998; 11(12): 1480 – 1485.