SERUM VITAMIN E, C AND A LEVEL IN LUNG CANCER: A CASE CONTROL STUDY

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Abstract

Lung cancer is a leading cause of cancer death worldwide, causing approximately 1.2 million deaths per year. Lung cancer is prevalent in smokers. It is documented that smoking induces oxidative stress, which is thought to be associated with the aetiology of carcinogenesis. A case control cross sectional study was conducted on sixty lung cancer patients and sixty smokers as cohort control. Serum levels of antioxidant vitamin E, C and A among the case and control subjects were estimated. It was observed that vitamin E value among the study population did not alter (15.67±3.67µmol/L vs. 14.66±3.88µmol/L). It was within normal range. Conversely, there was significantly high serum concentrations of vitamin C and A in the lung cancer patients as compared to those in the smoker controls (48.26±6.81versus 16.65±4.46µmol/L; 2.76±0.32 versus 1.60±0.35µmol/L respectively).

Keywords: Lung cancer, vitamin E, C and A, case control study

Introduction

Lung cancer is the leading cancer in male comprising 17% of the total new cancer cases and 23% of the total cancer deaths in the world (Jemal *et al.,* 2011). In Bangladesh it is also the most common cancer of males (Akhtar *et el.,* 2011). Epidemiologic evidence indicates that diets high in carotenoid-rich fruits and vegetables, as well as high serum levels of vitamin E and beta carotene, are associated with a reduced risk of lung cancer (The Alpha-Tocopherol Beta Carotene Cancer Prevention Study Group, 1994, Lee

et al., 2003, Padayatty et al., 2006). However, several large scale studies with intervention of β-carotene alone or combination of β-carotene and α-tocopherol unexpectedly could not demonstrate prevention of lung or breast cancer (Lotan, 1999, Albanes, 1999). Even some study concluded that intake of β-carotene or α-tocopherol may increase the risk of lung cancer in the smokers (Lotan, 1999, Edelson, 2008). However, there is overwhelming evidence that a high intake of vitamin C correlates with a low risk for cancer. Vitamin C helps prevention and protection of cancer incidence, even mega doses of vitamin C could cure cancer (Ocké *et al.*, 1997, Padayatty *et al.*, 2006, Hickey *et al.*, 2008). The present study has attempted to address the serum vitamin E, C and A level in lung cancer patients in comparison with those in the smokers.

Materials and Methods

Study population

It is case control cross sectional study conducted on sixty lung cancer patients and sixty smokers as cohort controls of age between 40 to 60 years. The hospitalized lung cancer patients were randomly selected from Dhaka medical college hospital, Mitford hospital, Cancer institute and some clinics in Dhaka city. Patients were histologically diagnosed and on routine treatment and dietary supplementation with vitamin C. Age and socioeconomic matched smokers smoking 15 or more cigarette sticks/day for a period of 10-15 years or >15 years were purposively enrolled as cohort control from the community. Patients were in medication and dietary supplementation. Blood specimens was collected from each of the patients and control subjects

Serum analysis for estimation of vitamin E, A and C

A 5ml of venous blood sample was collected from antecubital vein from each of the patients and control subjects for analysis of vitamin E, C and A. Serum was extracted from the blood by keeping it undisturbed for 60 minutes and then centrifuged at 3000 rpm for 10 minutes. A reversed-phased HPLC (LC-10AD, Shimadzu, HPLC 1991, model-1725, Japan) was employed for simultaneous determination of vitamin E and A as describe by Islam *et al* (2001). The analytes vitamin E and A in the sera were isolated by liquid-liquid extraction using n-hexane and ethanol, which was concentrated by evaporation under nitrogen. A 50µl reconstituted analyte was injected into chromatography on a C^{18} shim pack CLC-ODS (M) column of diameter 4.6mm (Shimadzu, LC column, 4.6x250mm, no. 1256168, Japan) with methanol:water (95:5 ratio) mobile phase flowing at 1ml/min, detector set at one attenuation. Every sample was injected twice to obtain replicate

chromatographs. Standard analytes were injected for every 25 test samples. Vitamin E and A were detected spectrophotometrically at 291nm.

For vitamin C analysis, serum immediately after extraction from blood was treated with 5% trichloroacetic acid and centrifuged at 3000 rpm for 10 minutes. It was then treated phenyl hydrazine and absorbance was read against a reagent blank at 520nm in a Spectrophotometer (UV-1201, UV-vis, Shimadzu, Japan as describe by Islam *et al* (2001). Every sample was analyzed twice to obtain replicate readings. Standards (retinol, α-tocopherol, retinol acetate and α-tocopheryl acetate, ascorbic acid) were purchased from Sigma Chemical Co., USA and solvents (HPLC grade) were from Merck (Darmstadt, Germany).

Statistical analysis

The SPSS software package (12.5 SPSS version Inc. Chicago USA) was used for statistical analysis. Data were presented as mean±sd. Comparison of serum vitamin levels between groups was performed by student's *t* test. P<0.05 was considered as a level of significance.

Results and Discussion

Table 1 shows serum concentrations of vitamin E, C and A in the lung cancer patients and healthy smokers as control subjects. Vitamin E value among the study population was found to be within normal range (Young, 1998). Although it was higher in the lung cancer patients (15.67 ± 3.67µmol/L) than that in the smokers (14.66 ± 3.88µmol/L), the difference was insignificant. In case of vitamin C, it was significantly (p=0.000) high (48.26 ± 6.81µmol/L) in the lung cancer patients as compared to that in the smokers (48.26 ± 6.81versus 16.65 ± 4.46µmol/L; p=0.000). Lung cancer patients had have significantly (p=0.000) higher serum vitamin A level than that in the smokers (2.76 ± 0.32 versus 1.60 ± 0.35µmol/L).

Table 1: Serum vitamin E, C and A level in lung cancer patient and smoker control

Serum level (µmol/L)*	Lung cancer	Smoker control	Level of significance
Vitamin E	15.67 ± 3.67	14.66 ± 3.88	P=0.635
Vitamin C	48.26 ± 6.81	16.65 ± 4.46	P=0.000
Vitamin A	2.76 ± 0.32	1.60 ± 0.35	P=0.000

*value expressed as mean ± SD
student's *t* test: p<0.05 as level of significance

Lung cancer is a leading cause of cancer death worldwide, causing approximately 1.2 million deaths per year (van der Meij *et al.*,2012). Oxidative stress is thought to be associated with the etiology of carcinogenesis (Klaunig and Kamendulis, 2004, Toyokuni, 2006). Smoking induces oxidative stress (Carnevali *et al.*, 2003). It is also evident that lung

cancer is more prevalent among heavy smokers than the non-heavy smokers (Kumagai *et al.,* 1998). Antioxidants vitamins including β-carotene have been reported to reduce oxidative stress. They are inversely associated with the risk of development of various cancers, even could cure cancer (Ocké *et al.,* 1997, Lotan, 1999, Padayatty *et al.,* 2006, Hickey *et al.,* 2008, Chen et al., 2008). This study has made an attempt to address serum vitamin E, C and A in lung cancer patients in comparison with those in the smokers.

Results showed that there had no significant change in the serum vitamin E level between the lung cancer patients and the smokers. This finding is to some extent consistent with the report that serum antioxidant vitamins are not associated with risk of cancers (Katsoulis et al., 2007, Emri *et al.,* 2012). There is overwhelming evidence that a high intake of vitamin C correlates with a low risk for cancer (Block, 1991). Vitamin C is the strongest antioxidant, mopping up the dangerous free radicals. It has been used as a dietary supplement intended to prevent oxidative stress–mediated chronic diseases including cancer. Vitamin C helps prevent and treat cancer by enhancing the immune system; stimulating the formation of collagen necessary for 'walling off' tumours; preventing metastasis by inhibiting a particular enzyme and thus keeping the ground substance around tumours intact; preventing viruses causing cancer; correcting a vitamin C deficiency in cancer patients; enhancing the effectiveness and reducing the toxicity of some chemotherapy and preventing free radical damage and neutralising some carcinogens. Mega doses of vitamin C could cure cancer (Chen *et al.,* 2005, 2008). Cancer cells cannot survive in vitamin C rich environment. It is reported that vitamin C as an antioxidant reduces systemic oxidative stress, but as pro-oxidant selectively induces local oxidative stress in cancer cells and thus kills the cancer cells (Verrax et al., 2009). The vitamin C supplementation and high intake of fruits rich in vitamin C and carotenes would be the possible causes for higher serum vitamin C and A levels in sera of the lung cancer patients. Lower serum levels of vitamin C and A in the smokers may be because of oxidative stress induced by tobacco smoking (Carnevali et al., 2003, Aoshiba, 2003).

Conclusion

Serum vitamin E value among the study population did not alter, which was within normal range. However, serum concentrations of vitamin C and A in the lung cancer patients were significantly high as compared to those in the smoker controls.

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