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Prostatic Alteration of Serum Levels of Carcino-Embryonic Antigen, Zinc and Selenium among Patients with Benign Prostatic Hyperplasia and Cancer of the Prostate

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Abstract:

Background: *Prostate disorder is defined as any of the abnormalities that affect the prostate gland in the male reproductive system and mostly comprised of prostatitis, benign prostatic hyperplasia (BPH) and prostate cancer (CaP). Prostate disorders are becoming a major public health issue in Nigeria adult men with incidence rates of 11% for CaP, 25% for BPH and 12% for prostatitis. This study was designed to evaluate the prostatic alterations that may be associated with the serum levels of Zinc and Selenium in patients*

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with prostate disorders. ***Materials and methods:*** *A total number of 55 men diagnosed with cancer of the prostate (CaP), 52 men diagnosed with Benign Prostatic Hyperplasia (BPH) and 55 apparently healthy men attending the urology clinic in Nnamdi Azikiwe Teaching Hospital were recruited for this study. The serum levels of Carcinoembryonic antigen (CEA) was analysed by ELISA method while the serum levels of Zinc and Selenium were determined using Atomic Absorption Spectrophotometer (AAS). Inferential Statistical analysis was done using version 22 of statistical package for social sciences (SPSS).* ***Results:*** *The mean serum level of Zn was significantly lower (P<0.05) in BPH (0.41±0.16) and CaP (0.55±0.34) subjects compared with the control subjects (0.85±0.42). The mean levels of Zinc and Selenium were also significantly increased in CaP subjects (0.55±0.34 and 0.30±0.15) compared with BPH subjects (0.41±0.16 and 0.27±0.09) (P<0.05) respectively. However, the mean levels of CEA did not differ in BPH and CaP subjects.* ***Conclusion:*** *We therefore observed that the relatively low serum levels of Zinc and Selenium in both BPH and CaP subjects, may predispose the prostate-cell into oxidative stress, reduce their protective mechanisms and therefore promote the damaging effects associated with benign prostatic hyperplasia and cancer of the prostate.*

Key words: Prostate, Benign prostatic hyperplasia, Prostate cancer, Zinc, Selenium, Carcinoembryonic antigen.

INTRODUCTION:

Prostate disorders refer to disease conditions of the prostate commonly classified into three major prostate abnormalities namely; prostatitis, benign prostatic hyperplasia (BPH) and prostate cancer.[1] Prostatitis is defined as an increased number of inflammatory cells within the prostate gland. The inflammatory process may be infectious or inflammatory in origin.[2] It's a common condition which can affect men of any age, but it's most common in younger and middle aged men, typically between 30 and 50 years of age.[1]

Benign Prostatic Hyperplasia (BPH) is an age-related non-malignant enlargement of the prostate gland.[3] Anatomically, BPH is often associated with the posterior urethral glands (PUG) and transitional zone (TZ) of the prostate.[4] In men over the age of 60 years, enlargement (hyperplasia) of the prostate is relatively common. In the vast majority of cases it causes no symptomatic difficulties, though infection may occur, as may rupturing of blood vessels. Enlargement may cause compression of the urethra with progressive obstruction of the flow of urine, incomplete emptying, or inability to void; there may also be a constant dribbling of urine. The bladder is never totally emptied, however, and the remaining urine becomes stagnant and infection sets in. The stagnant urine may cause the precipitation of stones in the bladder; the bladder muscle thickens to overcome this obstruction.[4] If urine begins to back up in the kidney, progressive damage may ensue, which can lead to kidney failure and subsequent uremia (the toxic effects of kidney failure). In severe cases, BPH may lead to sepsis, irreversible bladder damage, renal failure or even death.[5]

Prostate cancer is a malignant (cancerous) tumour (growth) that consists of cells from the prostate gland. Prostate cancer is the most common malignant tumour in men over the age of 65 years.[4] Thus, prostate cancer under age 40 is extremely rare, while it is common in men older than 80 years of age. As a matter of fact, some studies have suggested that among men over 80, between 50 and 80 percent of them may have prostate cancer.[6] It has been declared a public health epidemic in black American men because of its high incidence.[7] Prostate cancer most commonly metastasizes to the bones, lymph nodes, and may invade rectum, bladder and lower ureters after local progression. Prostate cancer has the potential to advance loco-regionally to adjacent organs. This spread can take place via different routes, including direct invasion and through lymphatic channels.[8]

The trend at which prostate cancer is growing is alarming and therefore requires urgent attention. However, the current screening method for prostate cancer relies on a combination of Prostate specific antigen (PSA) assay and a Digital Rectal Examination (DRE) while biopsy is done to confirm it if there is suspicion of cancer. Thus, the interest of this study is to evaluate if some important trace elements (Zinc and Selenium) and CEA could be of diagnostic value.

Carcinoembryonic antigen (CEA) is an acute phase protein that is synthesized in the gastrointestinal tissue during fetal development, but the production stops before birth.[9] As a carcino-embryonic antigen, it is present in trace amount in healthy individuals but rise significantly following disease conditions. Therefore, its relative importance in disease states especially in cancerous conditions (carcinoembryonic antigen), it could be of good value in determination of prostate diseases.[10] Trace elements are inorganic molecules which are essential for life. Although these elements constitute a relatively small amount of total body tissues, they are very essential in many physiological and biochemical processes. In states of absolute deficiency, death results and in limited intake biological functions are impaired.[11] Zinc is an important constituent of prostatic fluid and has been postulated to play important roles in the development and normal functioning of prostate.[12,13] According to Huang *et al*. (2006), prostate tissues from healthy individuals are often associated with the highest levels of zinc as a result could be a good diagnostic importance in prostate disorders.[14]

Selenium is an essential trace element present in foods such as bread, cereals, nuts, meat, fish and other seafood. It is predominantly known as the amino acid derivatives, selenomethionine and selenocysteine.[15] There is a relatively narrow margin between selenium intakes that result in deficiency or toxicity, with health effects being related to level

of exposure and selenium status.[16] Without selenium, the function of the selenium-requiring proteins can be compromised which results in could be detrimental to health. Certain diseases such as cancer and cardiovascular disease as well as aging process, is associated with an increase in oxidative damage.[17] Maintaining adequate selenium intakes may provide some protection against these processes.

MATERIALS AND METHODS

This was a cross-sectional survey that was carried out in the Surgical Out-patients Department of Nnamdi Azikiwe University Teaching Hospital Nnewi (NAUTH). Ethical clearance was obtained from the Ethics Committee of Nnamdi Azikiwe University Teaching Hospital, Nnewi before the commencement of the study and informed consent was obtained from the subjects.

Study subjects and design

This study comprised of a total of 162 subjects which consist of 52 individuals with Benign Prostate hyperplasia, 55 subjects with Prostate cancer and 55 apparently healthy men that served as control group.

Sampling Technique

Random sampling technique was used during sample collection. 5 ml of whole blood was collected from subjects as they visited the Urology unit at Nnamdi Azikiwe University Teaching Hospital using a plain specimen container. The serum obtained after centrifugation was stored at 2-8°C until analyzed.

Inclusion criteria

Individuals within the age of 50-80 with Benign Prostate Hyperplasia or cancer of the Prostate were recruited.

Exclusion criteria

Subjects that have disease conditions such as alchoholic liver disease, liver cirrhosis were excluded from this study.

Statistical analysis

The version 22 of Statistical Package for Social Sciences (SPSS) was used in statistical analysis. The results were expressed as (Mean±SD). Comparisons were made using one way analysis of variance (ANOVA). Pearson correlation analysis was used to establish possible correlation between analytical parameters and BHP, CaP and Prostatitis. $P<0.05$ was taken as significant.

METHODS OF ANALYSIS:

Determination of CEA

The method of Gold and Freedman (1965) was adopted. This method is an Enzyme-Linked Immunosorbent Assay procedure.

Principle:

The CEA ELISA test is based on the principle of a solid phase enzyme-linked immunosorbent assay. The assay system utilizes a monoclonal anti-CEA antibody for solid phase (microtiter wells) immobilization and another mouse monoclonal anti-CEA antibody in the antibody-enzyme (horseradish peroxidase) conjugate solution. The standards and test specimen (serum) are added to the CEA antibody coated microtitre wells. Then CEA antibody labelled with horseradish peroxidase (conjugate) is added. If human CEA is present in the specimen, it will combine with antibody on the well and the enzyme conjugate resulting in the CEA molecules being sandwiched between the solid phase and enzyme-linked antibodies. The concentration of CEA is directly proportional to the color intensity of the test sample.

Measurement of Zinc and Selenium:

The working principle of Atomic Absorption Spectrophotometer is based on the sample being aspirated into the flame and atomized when the light beam of the AAS is directed through the flame into the monochromator, and onto the detector that measures the amount of light absorbed by the atomized element in the flame. Since metals have their own characteristic absorption wavelength, a source lamp composed of that element is used, making the method relatively free from spectra or radiational interferences. The amount of energy of the characteristic wavelength absorbed in the flame is proportional to the concentration of the element in the sample.

RESULTS:

Anthropometric data of subjects with prostate disorder and control subjects

The height, weight and body mass index (BMI) of the BPH, CaP and control subjects are shown in table 4.1. The result showed that there was no significant differences (P>0.05) in weight (72.56±10.97, 75.85±12.87 and 75.79±11.94), height (1.68±0.06, 1.70±0.08 and 1.71±0.08) and BMI (25.55±3.54, 26.11±3.27 and 25.92±3.13) of the BPH, CaP and control subjects respectively.

Table 1: (Mean ± SD) of the anthropometric data of subjects with prostate disorder and control subjects.

Parameter	BPH n=52	CaP n=55	CONTROL n=55	F value	*P* value
Weight (kg)	72.56±10.97	75.85±12.87	75.79±11.94	1.310	0.349
Height (m)	1.68±0.06	1.70±0.08	1.71±0.08	1.432	0.227
BMI (kg/m^2)	25.55±3.54	26.11±3.27	25.92±3.13	0.399	0.374

KEYS: BMI= Body Mass Index, CEA = Carcino-Embryonic Antigen, BPH = Benign Prostatic Hyperplasia, CaP = Cancer of the Prostate. Mean difference is significant when P is <0.05. * = mild significance and ** = marked significance.

Variations of the serum levels of CEA, Zinc and Selenium in BPH, CaP and Control Subjects.

The results showed that the mean levels of zinc and selenium were significantly higher in CaP subjects (0.55±0.34 and 0.30±0.15) compared with BPH subjects (0.41±0.16 and 0.27±0.09) ($P<0.05$) respectively. Similarly, the serum levels of zinc and selenium were significantly lower in BPH (0.41±0.16 and 0.27±0.09) and CaP (0.55±0.34 and 0.30±0.15) subjects when compared with control subjects (0.85±0.42 and 0.65±0.59). However, the mean level of CEA did not show any significant difference in BPH (0.18±0.06), CaP (0.23±0.18) and control groups (0.21±0.09) ($P>0.05$).

Table 2: Variations of the serum levels of CEA, Zinc and Selenium in BPH, CaP and Control Subjects.

Parameter	BPH n=52	CaP n=55	Control n=55	F-value	P-value
CEA (ng/ml)	0.18±0.06	0.23±0.18	0.21±0.09	2.759	0.066
Zinc	0.41±0.16[a,b]	0.55±0.34[a,b]	0.85±0.42[a]	25.257	0.001*
Selenium	0.27±0.09[a,b]	0.30±0.15[a,b]	0.65±0.59[a]	19.446	0.000**

KEYS: CEA = Carcino-Embryonic Antigen, BPH = Benign Prostatic Hyperplasia, CaP = Cancer of the Prostate. Mean difference is significant when P is <0.05. * = mild significance and ** = marked significance. a = significant difference between BPH, CaP and control, b = significant difference between BPH and CaP only.

DISCUSSION:

The incidence of benign prostate hyperplasia and prostate cancer is rapidly increasing worldwide. However, drastic attention is needed in order to reduce the rate of mortality and morbidity due to these prostate diseases.

In the present study, the mean carcinoembryonic antigen (CEA) level showed no significant difference in BPH and prostate cancer subjects compared with control. This was similar to a previous study which reported that there was no correlation between CEA and PSA.[18] For so many years CEA has been used as tumor marker in different carcinomas, but it

is not known whether it can be useful in prostate cancer. This may suggest that CEA is mostly a marker for colorectal, gastrointestinal and breast cancer and not for prostate. CEA functions as a differentiation inhibitor and cell adhesion molecule, in vitro. Thus, may be of good value in prostatic glandular lesions.[19]

More so, the significant decrease in the mean levels of zinc and selenium observed in subjects with prostate cancer and BPH subjects compared with the control group suggest that plasma and tissue zinc and selenium are substantially lowered in cancerous prostate and BPH than in normal prostate.[20,21,22,13]. This is because, total zinc levels in the prostate are much higher than in other soft tissues in the body, and those with prostate cancer have been shown to have exceedingly low levels of zinc in the prostate.[23,13] and prostatic secretions[23] in men with prostate disease compared with healthy men. Nevertheless it was suggested that zinc deficiency could be a risk factor for prostate cancer.[24] It is well documented that tumor cells undergo metabolic transformations that are essential for their malignant existence but are not the cause of malignancy. The accumulation of zinc in normal prostate glandular epithelial cells results in two important effects, a metabolic effect, and a proliferative effect. Its metabolic effect is the inhibition of citrate oxidation, which is necessary for prostate function. A second effect of zinc is its inhibition on prostate-cell proliferation.[25] Therefore, Zinc is of utmost importance for the functions of many transcriptional factors and proteins involved in the recognition of specific DNA sequences and regulation of gene transcription. Zinc also has a protective effect against the free-radical injury.[26] As a result may also play a key role in the prevention of prostatic disease by ameliorating oxidative stress, which can subsequently result in DNA damage, increasing the risk of mutation and malignant transformation. In fact, dietary Zn deficiency has been

associated with increased DNA damage in the prostate during oxidative stress.[27] Specifically, Zn deficient prostate cells have greater DNA damage and altered expression of genes associated with this damage, indicating that marginal Zn intake may sensitize the prostate to oxidative damage. As oxidative stress increases, so does the cellular Zn requirement for protective mechanisms, thus perpetuating the harmful effects of Zn deficiency.[28,27] According to Liang et al. (1999), the inhibitory effect of zinc on human prostatic carcinoma cell growth, may be possibly due to the induction of cell cycle arrest and apoptosis.[29] Costello et al. (1997) reported that zinc inhibits mitochondrial aconitase and exerts its importance in the citrate metabolism of prostatic epithelial cells.[25] The intra-mitochondrial accumulation of high zinc levels inhibits m-aconitase activity which inhibits citrate oxidation. This essentially truncates the Kreb's cycle and markedly decreases ATP production (normally coupled to citrate oxidation). These relationships form the basis of a new concept of the role of zinc and citrate related energy metabolism in prostate malignancy. The inability of carcinoma cells to accumulate high zinc levels results in increased citrate oxidation and the coupled ATP products essential for progression of malignancy.[29] Feng *et al.* (2000) showed similar effects of high intracellular accumulation levels of zinc in prostate cells inducing mitochondrial apoptogenesis.[30]

Furthermore, it has also been discovered that higher serum selenium levels may reduce prostate cancer risk in men with high intake of vitamin E, in multivitamin users, and in smokers.[31] Interventions with selenium have shown great benefits in reducing the risk of cancer incidence and mortality in all cancers combined, and specifically in liver, prostate, colo-rectal and lung cancer.[15] These findings are in agreement with two meta-analyses of observational studies from diverse populations that reported the potential effect of selenium

against the development of prostate cancer.[21,22] In contrast to these findings, certain studies observed that there was no significant difference in mean selenium levels in BPH and CaP subjects.[33,32,31] Goodman *et al.* (2001) carried out a study on 235 prostate cancer cases and 456 controls and found no association between prostate cancer risk and serum selenium concentrations in a cohort from the Carotene and Retinol Efficacy Trial.[33] In 2004, Lipsky *et al.* evaluated 150 participants (70 cases and 80 controls) and reported toe nail selenium levels may not influence prostate cancer incidence.[34] Similarly, Allen and colleagues (2004) suggested that selenium concentration, as measured in nails of 300 case control pairs, did not strongly associate with prostate cancer risk.[32] Peters *et al.* (2007) observed no inverse association between pre-diagnostic serum selenium concentration and the risk of prostate cancer in a large cohort study with 724 cases and 879 matched controls.[31] Prostate cancer risk is an age-related disease and oxidative stress increases with age,[35] through generation of reactive oxygen species and reduced activity of anti-oxidative enzymes.

CONCLUSION:

We therefore conclude that the relatively low serum levels of Zinc and Selenium in both BPH and CaP subjects, may predispose the prostate-cell into oxidative stress, reduce their protective mechanisms and therefore promote the damaging effects associated with benign prostatic hyperplasia and cancer of the prostate. This is substantiated by the fact that the prostate gland is androgen sensitive hence; the prostate may be susceptible to oxidative damage because androgens increase oxidative stress, partly by increasing the mitochondrial and decreasing the glutathione activities.

The serum levels of Zinc and Selenium estimation can be an adjunct in the identification of adult men at risk for development of prostatic complications such as BPH and CaP. Zinc and Selenium supplementation should also be encouraged in the management of prostate disorders so as to reduce oxidative stress associated with reduced levels of zinc and selenium.

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