Prospective Study of Sex Hormone Levels among Prostate Cancer Patients Attending the University of Port Harcourt Teaching Hospital Clinic.

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ABSTRACT

Background: Longstanding and diverse body of evidence supports the view that sex steroids play a role in the development of prostate cancer. Epidemiological and demographic studies in humans as well as animal experiments have sought to determine the independent effect on risk as well as the interrelationship between these hormones. In this study, we investigated the variations in testosterone and oestradiol levels among prostate cancer patients attending UPTH clinics with the objective of determining the role played by sex hormone variations in the development of cancer of prostate in patients attending the University of Port Harcourt Teaching, Port Harcourt.

Methods: Newly diagnosed patients with cancer of the prostate attending the Urology clinic of University of Port Harcourt Teaching, Port Harcourt from December 2011 to April 2012 were recruited for the study. Their Prostate Specific Antigen (PSA) testosterone and oestrogens levels were measured using Elisa Kits. Correlation between individual hormone levels in control subjects were assessed by Spearman correlation coefficients (R). Student t-test was used to assess if there was any significant difference between the patients and controls in the level of these hormones. We computed the ratio of oestradiol to testosterone and compared case patients with control subjects by use of t-test at 95% confidence interval. Test cases were also divided into two groups by age to study variations across subgroups.

Results: 105 patients recently diagnosed with prostate cancer and 40 normal subjects were analyzed. We observed a negative correlation between testosterone and oestradiol ($r = -0.66$). Testosterone and oestradiol levels in prostate cancer patients were also significantly different from that of controls. Mean testosterone level in control was 3.2 ng/ml while that of the patients was 4.0 ng /ml. Mean oestradiol level in controls was 32.8 pg/ml while that of the patients was 21.2pg/ml ($p < 0.05$ in both cases). The ratio of oestradiol to testosterone was also significantly altered in prostate cancer patients ($p < 0.05$). The mean levels of hormones and hormone ratios across the two sub-age groups were not substantially different in patients with prostate cancer ($p > 0.05$).

Conclusion: This study indicates that increased levels of testosterone in circulation are associated with risk of prostate cancer. This risk is further associated with low levels of circulating oestradiol. The relative levels or ratio of these hormones are very important in the development of prostate cancer. Age of the patient appear not to be strongly related with these changes after the cancerous state has set in.

Key Words: Prostate cancer, oestrogens, testosterone, Prostate Specific Antigen (PSA).

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INTRODUCTION

Prostate cancer was the second most frequently diagnosed cancer and the sixth leading cause of cancer related death in males worldwide as at 2011[1]. In Nigeria, 2% of men develop cancer of the prostate with a prevalence of 127 per 100,000[2]. It has long been documented that sex steroids, particularly androgens play a role in the pathogenesis of prostate cancer[3]. Data from animal experiments and from epidemiological and endocrinological studies in humans supports the evidence that the individual hormones which control normal growth of target organs can also create the proper conditions for endoplasmic transformation. Androgens are essential for normal growth and maintenance of the prostate gland; however it has also been proven from studies to stimulate the proliferation of human prostate cancer cells in vitro and can indeed cause prostate cancer in rodents when given in large quantities [4,5].

Androgen deprivation frequently causes prostate cancer to regress and is a common approach in prostate cancer therapy. Additionally it is known that eunuchs rarely develop prostate cancer[6] (Huggins and Hodges, 1941). On the other hand, estrogen therapy has a palliative effect in advanced prostate cancer and a reduced risk of prostate cancer has been associated with certain hyper-estrogenic states .[3] Gann and his co-workers in their study particularly observed an association between testosterone and prostate cancer. The association with estrogen was though weaker. These observations add credibility to the well-known fact that extreme variations in sex hormone levels constitute a risk and indeed play a role in the etiology of prostate cancer[7]. As far back as 1948, there was already evidence suggesting that exogenous estrogen causes extreme degeneration in the peripheral zone of the prostate where most cancers arise but very little in the central zone[8].

Studies on the role of sex steroids in prostate cancer have been done in the Niger delta region of Nigeria seem to be uncommon. We have therefore assessed the androgen and estrogen hormone levels in prostate cancer patients attending the Urology clinic of the University of Port Harcourt Teaching Hospital. Our focus was on testosterone and estrogen, the major contributors of androgenic and estrogenic activities in the body respectively.

METHODS

Study Design and Population: This was a cross sectional prospective study comparative study. Samples were collected from patients from the surgical outpatient and urology clinics of the University of Port Harcourt Teaching Hospital who came to the Hormone Laboratory of the Department of Chemical Pathology to do their PSA tests from December 2011 and April 2012.

Inclusion criteria for the subjects were a recent diagnosis of histologically confirmed prostate cancer irrespective of the stage. Patients who had already initiated therapy or who have had an orchidectomy and or prostatectomy were excluded from the study.

Control subjects were selected from cases that had come to the Hormone Lab for routine PSA screening. Those selected as controls were those who had their PSA levels falling within the normal range (0-4 ng/ml), and who had no previous history of prostatectomy and were not on any known drugs for prostate disease.

Sample Collection: Blood samples from patients that met the criteria for the study were collected into plain sample bottles by venipuncture at the phlebotomy unit of Chemical Pathology Department. Serum obtained from the samples after centrifugation at 2,000g was stored in a frozen state for not more than 2 weeks before analysis.

Laboratory Assays: Previously unthawed serum samples for case patients and controls were analyzed at the Hormone Lab of Chemical Pathology Department of the University of Port Harcourt Teaching, Port Harcourt. Samples were run in batches and in duplicates. PSA, testosterone and estradiol were measured by use of Enzyme Linked Immunoassay kits obtained from BioCheck (Vintage Park Dr., Foster City, CA).

Data Analysis: Data were analyzed for each group of sample to obtain the mean, standard deviation and coefficient of variation for control and test subjects for the different parameters. The student t-test was used to determine significance of difference in mean of the two groups at 95% confidence interval. Correlations between hormone levels were evaluated using the control data only by calculation of Spearman correlation coefficient.

We assessed for trends or variation across sub-groups by dividing the test group into two sub-groups by age. We also computed the ratio of estradiol to testosterone, a ratio that has been construed as an index of hormone balance or metabolism and compared the test patients with control subjects to see if there was any significant difference.

Ethical Clearance: Ethical approval was sought from the University of Port Harcourt Teaching Hospital, ethics and research committee and all patients gave informed consent to participate in the study.

RESULTS

One hundred and five patients (105) patients were selected for the study and forty (40) control subjects were tested. As shown in table 1, the mean age of the study group was 71.8 years while it is 54.2 years for the controls ($p < 0.5$).

The mean PSA level was 60.7 ng/ml for the study group and 1.3ng/ml for the controls ($p<0.05$). The mean testosterone level of the patients was 4.0ng/ml. This was significantly different from that of the control subjects (3.2ng/ml), $p<0.05$. The mean values of oestradiol in patient group was also significantly different from that of the control group; 21.2pg/ml and 32.8pg/ml respectively ($P<0.05$). The oestradiol to testosterone ratio (E2/T) in patient group was 5.3, while ratio was 10.7 in the control group. This was significantly different ($p<0.05$). The mean values for the testosterone and oestradiol as well as their ratios were also shown in table 1. The P values and results of Students''t' test at 95% confidence interval were recorded as well.

Table 2 shows the mean values of the selected parameters for the test group subdivided into two groups by age. The mean levels of hormones and hormone ratios are not substantially different in the 2 subgroups of patients already diagnosed with prostate cancer. There was no significant difference in the parameters in the case patients below and above the ages of 70 years.

There was a significant negative correlation for the association between estradiol and testosterone among control subjects. The correlation Coefficient(R) was -0.66 with $P>0.05$ as shown in Table 3.

Table1. Mean levels of selected parameters for prostate cancer patients and control subjects

	Case patients (n = 105)	Control Subjects (n = 40)	p-value	Significance
Mean age (Yrs)	71.8	54.2	4.16	P<0.5
PSA, ng/ml	60.7	1.3	5.51	P<0.05
Testosterone, ng/ml	4.0	3.2	2.44	P<0.05
Estradiol, pg/ml	21.2	32.8	2.90	P<0.05
Hormone Ratio, E2/T	5.3	10.7	2.73	P<0.05

Table2. Mean levels for selected parameters in case patients divided into 2 groups by age

	Age (yrs): 60-70 (n = 40)	Age (yrs): 71+ (n = 65)	P value	Significance
PSA ng/dl	61.8	60.0	0.77	p>0.05
Testosterone ng/ml	4.0	4.0	0.97	p>0.05
Estradiol pg/ml	19.5	21.7	0.13	p>0.05
Hormone Ratio E2/T	4.9	5.5	0.14	p>0.05

Table3. Association between estradiol and testosterone in control subjects. (n=40)

	Testosterone
Estradiol	R = -0.66 (P >0.05)

DISCUSSION

The mean age of our prostate cancer patients was 71.8 years. This is in keeping with other studies in Nigeria and USA.[1, 9]. This further confirms the known fact that old age is one of the major known risk factors for development of prostate cancer.

In this study, significantly higher levels of testosterone-within the normal endogenous range, were observed in prostate cancer patients implicating a role of testosterone in prostatic cancer. Testosterone levels were higher in case patients as compared with normal subjects. On the other hand, low levels of estradiol were associated with prostate cancer patients. These findings are in line with previous studies which suggests that the primary mechanism involved in prostate cancer pathogenesis might probably depend more on modification of the factors which affect secretion and metabolism of the responsible hormone rather than on exposure to classical exogenous initiators[4,10-13]. Testosterone is converted to dihydrotestosterone, which in turn supports the growth of the prostate. This higher level of testosterone thus translates to a greater possibility of proliferation of prostatic cells. It is therefore not surprising that we observed higher levels of testosterone for the prostate cancer patients when compared with normal controls.

We observed a negative correlation between testosterone and estradiol. This is at variance with the findings of Gann [14]et al (1996), but does agree with other findings[3]. As was stated earlier, a reduced risk of cancer is associated with certain hyper-estrogenic states and estrogen therapy has a palliative effect in advanced states[3, 6]. It does appear that the metabolic balance between estrogen and testosterone is altered in the cancerous state indicating that the ratio of these hormones rather than their absolute values may be more important in the pathogenesis of prostate cancer. We observed a significant difference in the ratios of estrogen to testosterone in prostate cancer patients ($p<0.05$).

We divided the case study patients into 2 groups by age 60-70 years and 71 years and above, in order to see if age had any effect on these outcomes. From our results (Table2), there was no significant difference in the mean level of estrogen and testosterone in the different age groups. There was a slight difference in the mean of the ratio of estrogen to testosterone but this was not significant suggesting again that the ratio of these hormones rather than their absolute values are implicated in prostate cancer etiology. Though the risk of prostate cancers and benign hypertrophy increases with age, it does appear that after the cancerous state has set in, these ratios rather than other factors play a greater role in creating the favorable environment for these cancerous cells to proliferate[5].

The result of this study does support the theory that long term exposure to modifications in the levels of endogenous testosterone and estradiol is implicated in prostate cancer development. In this study we did not include sex hormone binding globulin (SHBG). The outcome of this study may have been different if this had been measured, as there is the hypothesis that only the bioavailable hormone and not protein bound component is free to enter into cells to initiate action[15]. This is one limitation of this study. Forty four percent of all circulating testosterone is bound to SHBG, 2% is free and the rest is loosely bound to albumin and can enter cells to mediate an effect. Similar distribution applies to estrogen though it has a weaker binding affinity for SHBG. It follows therefore that increases or decreases in SHBG levels would affect bioavailability of both testosterone and estrogen[16].

The Levels of testosterone, estrogen and SHBG could have overlapping or distinct set of effects on prostate cancer. Investigation of what independent or confounding effect on risk hormones may have and modification of these levels with the goal of developing models for prostate cancer prevention or therapy should guide further research.

CONCLUSION

This study indicates that increased levels of testosterone in circulation are associated with the risk of developing prostate cancer. This risk is further associated with low levels of circulating oestradiol. The relative levels or ratio of these hormones are very important in the development of prostate cancer. Age of the patient appear not to be strongly related with these changes after the cancerous state has set in.

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