An Association Between Human Epidermal Growth Factor Receptor 2 and Prostate Cancer

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Abstract

Background: Prostate cancer is a momentous health problem worldwide. Prostate cancer in Sudan is the third most common cancer type. The role of human epidermal growth factor receptor 2 (HER2/neu) in prostatic tumors is still not fully understood. The findings of studies testing the association between HER2/neu expression and prostate cancer have been inconsistent.

Objectives: To demonstrate the association between ages, epidermal growth factor receptor 2 and prostate cancer.

Design: Retrospective.

Settings: Sudan University for Science and Technology and National Health Laboratory (NHL) in Khartoum state Republic of Sudan.

Patients and methods: Two paraffin sections were taken, one stained with Haematoxylin and Eosin to confirm the diagnosis, the other stained using primary antibodies to HER2 protein with immunohistochemistry method using the DAKO Hercep Test™ protocol.

Sample size: Forty-six paraffin blocks were included in this study; 36 specimens were medium-grade and high-grade cancers and ten specimens were low-grade cancer.

Results: The mean age of the study group was 64 years. Prostatic cancer grades revealed, 10 (21.7%) patients with Gleason Group = 3 + 3, 2 (4.3%) patients with Gleason Group = 3 + 4, 11 (23.9%) patients with Gleason Group = 4 + 3, and 23 (50%) patients with Gleason Group = 4 + 4. Positive Her2 was found to be associated with high-grade (GG8) and medium-grade (GG7) compared to low-grade prostatic cancer (GG6).

Conclusions: The current study was a clue for a possible association between HER2/neu and prostatic cancer, and further studies might elucidate this connection.

Keywords: HER2/neu, Prostate Cancer

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androgen independence and in identifying patients more likely to present disease progression\(^ 15\). The aim of the present study was to demonstrate the association between ages, HER-2 and prostate neoplasia.

**Materials and methods**

This study was conducted at the Faculty of Medical Laboratory Sciences, histopathology department, Sudan University for Science and Technology and National Health Laboratory (NHL) in Khartoum state Republic of Sudan (May 2016 to June 2018). Paraffin wax sections were selected from patients that were previously diagnosed as prostatic cancer at the National Health Laboratory. Clinic pathologic data of patients were collected from the records of the archive of the National Health Laboratory; their ages were ranged from 40 to 90 years. Tiny biopsies and the focal nodular prostate cancer were excluded. Forty-six paraffin blocks were included in this study; Prostatic cancer classified in adenocarcinomas according to WHO 2016 each lesion was evaluated according to the Gleason Group (GG) classification\(^ 15\). Thirty-six specimens were medium-grade and high-grade cancers and ten specimens were low-grade cancer. The tissues were cut into 4-μm thick sections and mounted on microscope slides. All sections were incubated in an oven at 60°C for 1 h, then in three changes of xylene for 2 min each to remove the wax, and then treated with three changes of absolute ethyl alcohol each for 2 min, then rehydrated through gradual ethyl alcohol and placed in water. Two sections were taken from all cases; one section was stained with Haematoxylin and Eosin stain as described by Fischer\(^ 16\) to confirm the diagnosis.

**HER2 immunohistochemistry (IHC) was performed using the DAKO Hercep Test**\(^ 14\); The slides were boiled at low power in a domestic microwave oven in citrate buffer (pH 6) for 30 min, and cooled at room temperature for 20 min. Next, slides were transferred to distilled water and washed in phosphate buffer saline (PBS), then blocked in 0.5% hydrogen peroxide in methanol for 10 min, and next washed twice with phosphate buffer saline (pH 7.4) for 2 min each. Next, protein block serum-free was added to each section for 10 min, then incubated in primary antibodies to HER2 protein for 1 h, washed twice with phosphate buffer, followed by incubation in biotinylated secondary antibody for 20 min, then washed with PBS. Next, drops of streptavidin reagent were applied for 20 min, sections were washed in PBS, and 3,3′diaminobenzidine tetrahydrochloride (DAB) substrate chromogen was applied for 5 min. The sections were then washed in distilled water, stained with Mayer’s hematoxylin for 1 min, and washed in distilled water. Finally, sections were dehydrated in alcohol, cleared in xylene and mounted in DPX. All sections were stained in the same batch to eliminate inter batch variation. The slides were studied by a pathologist who remained blind about the clinical characteristics and histopathology on the arms of the study. HER2 expression was classified according to the modified DAKO criteria as follows: Negative (scores 0+/+), and positive (score 2+ & 3+), the cutoff for score 3+ are more than 10% strongly positive cells. According to Kolla et al.\(^ 17\), sections with scores of 2 and 3 were considered positive for HER2/neu.

**Statistical analysis**

Data analysis was carried out with SPSS version 16.0. The correlation between prostatic tumor and HER-2 expressions status was determined by the Chi-square test. P-values < 0.05 were considered statistically significant.

**Results**

The mean age of the study group was 64 years (median, 65 years; range, 40 to 90 years). In regards to Gleason Group (GG) classification, a total of 46 patients with a clinical and pathologic diagnosis of prostatic cancer showed 10 (21.8%) patients with GG = 3 + 3, 13 (28.2%) patients with GG = 4 + 3, and 23 (50%) patients with GG = 4 + 4. The grades of prostate tumors and age groups are presented in Table 1.

The staining intensity of HER2-positive expression in the paraffin section was compared between high-grade (GG8), medium-grade (GG7), and low-grade prostatic cancer (GG6). Table 2 shows the comparison between the detailed staining results of HER2 positive expression in different grades of prostatic adenocarcinoma among the study group. The significant differences in HER2-positive expression staining intensity were observed in high-grade (GG8) prostatic adenocarcinoma. However, the majority of low-grade prostatic cancer (GG6) cases showed positivity see illustrations (Fig. 1). Positive expression was seen in 84.8% of prostatic cancer cases (39

| Age groups | 40 – 50 | 51 – 60 | 61 - 70 | 71 - 80 | 81 - 90 | Total |
|------------|---------|---------|---------|---------|---------|-------|
| Gleason Group (GG) | No (%) | No (%) | No (%) | No (%) | No (%) | No (%) |
| (GG = 6) | 2 (4.3%) | 4 (8.7%) | 3 (6.6%) | 1 (2.2%) | 0 (0%) | 10 (21.8%) |
| (GG = 7) | 1 (2.2%) | 0 (0%) | 9 (19.5%) | 3 (6.5%) | 0 (0%) | 13 (28.2%) |
| (GG = 8) | 1 (2.2%) | 4 (8.7%) | 11 (23.9%) | 5 (10.9%) | 2 (4.3%) | 23 (50%) |
| Total | 4 (8.7%) | 8 (17.4%) | 23 (50%) | 9 (19.6%) | 2 (4.3%) | 46 (100%) |
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Table 2. HER2 expression and prostatic tumor grade

| Prostatic tumor grad (GG = 6) | (GG = 7) | (GG = 8) | Total |
|------------------------------|----------|----------|-------|
| HER2 expression classes      | No (%)   | No (%)   | No (%) | No (%) |
| Negative                     | 1 (2.2%) | 3 (6.5%) | 3 (6.5%) | 7 (15.2%) |
| Positive                     | 9 (19.6%) | 10 (21.7%) | 20 (43.5%) | 39 (84.8%) |
| Total                        | 10 (21.8%) | 13 (28.2%) | 23 (50.0%) | 46 (100%) |

P: Value with chi-square test

| P (GG = 6) | P (GG = 7) | P (GG = 8) |
|------------|------------|------------|
| 0.000      | 0.000      | 0.000      |

Negative (scores 0/1+), and positive (score 2+ and more), the cutoff for score ≥ 2+ is more than 10% strongly positive cells. According to Kolla et al., 2008.

![Microscopic findings](image)

Discussion

Prostate cancer is a momentous health problem worldwide. Prostate cancer in Sudan is the third most common cause of cancer-related deaths in men. This study confirmed the previous finding that the possibility of prostate cancer increases continuously with age. The median age in our study group was 65 years, with the highest percentages of more than 60% of the study group reported with high-grade (GG8) and medium-grade (GG7) prostatic cancer among those of 61–70 years old and older, the comparative risk for prostate cancer equivalent showed relations with age. Prostate cancer has long been recognized as a disease of older men. Nevertheless, the inclinations in the age-specific incidence rate of prostate cancer had distinctive patterns relative to geographic and racial deviation. In most Western countries, including North America and Western Europe, the current age-specific occurrence rate peaked in the 65–74-year-old group, and dropped beyond the age of 75 years. In our investigation, the high rates occurred through the 60s and older, with an increased incidence of moderate- and high-grade prostate cancer rise in the late 60s and over 70s in comparison with that in the 40–60 age groups. Despite current progress in prostate cancer investiga-
tion, the role of human epidermal growth factor receptor 2 (HER2/neu) in androgen-stimulated proliferation is still not fully understood. Furthermore, the investigation of the association between HER2/neu expression and prostate cancer is inconsistent. Generally, HER2 overexpression has been determined only by gene amplification and because of this, protein levels of HER2 have not been comprehensively explored and described in prostate cancer. The predictive importance of HER2 in breast cancer is currently well-known. Controversy remains about the significance of HER2 in prostate cancer. Therefore, in the present study, we investigated the association between HER2/neu and prostate cancer. HER2 positive expression was noticed in 84.8% of the prostatic cancer, with statistical significance (p < 0.001). Positive Her2 is found to be associated with high-grade (GG8), medium-grade (GG7), compared to low-grade prostatic cancer (GG6) in 43.5%, 21.7%, and 19.6%, respectively. Former studies of HER2 overexpression in numerous cancers have described varieties from 0% to 100% in immunohistochemical studies. The histopathological findings from this study demonstrated that the expression of HER2 protein is associated with high-grade prostate cancer, in which HER2/neu positive expression was seen in more than 84% of prostatic cancer cells. These results agree with Day et al., who found that HER2 expression is elevated in bone metastases of prostate cancer. Furthermore, our results differed from data reported by Sanchez and colleagues and Jorda and colleagues, both of which described a low level of HER2 expression in primary prostate tumors. Other researches have reported higher HER2 expression levels. These inconsistent sets of records are possible due to the use of diverse immunohistochemistry tests and antibodies. This would propose that levels of HER2 expression in prostate cancer are actually considerably lower than detected in breast and other tumor types.

Furthermore, HER2/neu shows a major role in appr-eciative the oncogenesis of prostate adenocarcinoma. For this purpose, illuminating the HER2/neu expression is principally important in androgen independent prostate cancer, due to the increasing interest in using anti-HER2 targeted therapies for progressive disease treatment. The current study hints at an association between HER2/neu and prostatic cancer, and more studies might elucidate this connection. The cases involved in our study were restricted, and would need long-term follow-up, but our present research has only been undertaken for only a few years. Also, it would be beneficial to include more grades of malignant tumors as well as ‘ benign’ groups.

Conclusions

There is no clear consensus on the ideal management of prostate cancer, particularly in aging men. Cautious se-

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