Epidermal Growth Factor Receptor Gene in Prostate Cancer after Radical Prostatectomy

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Abstract

Background: The aim of this study was to evaluate over expression of epidermal growth factor receptor (EGFR) gene in localized prostate cancers and determine its relation with clinical and pathological factors affecting the progression of the disease.

Methods: In the tissue samples of the patients with prostate cancer undergoing radical prostatectomy, EGFR expression was evaluated by immunohistochemistry technique. Those with scores 2 and 3 were considered to be positive for the expression. The relation between gene expression and grade was evaluated by chi-square test.

Results: Of 100 tissue samples evaluated, 80 and 20 had Gleason score (GS) < 7 and ≥ 7, respectively. Also, 68 and 32 had PSA level ≤ 10 and >10, respectively. A total of 59 samples were positive for EGFR expression, of whom, 46 had GS < 7 and 13 had GS > 7 (P-value = 0.39). Forty patients had PSA < 10 and 19 had PSA > 10 (P-value = 0.5).

Conclusion: Fifty-nine percent of the patients with localized prostate cancer undergoing radical prostatectomy were EGFR positive. No significant relation was found between EGFR and grading (GS) or PSA.

Keywords: Epidermal growth factor receptor; Gene expression; Prostate cancer

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Introduction

Radical prostatectomy or radiotherapy is a curative treatment for localized prostate cancer. Pathologic grading of the prostate cancer by Gleason Score (GS) is the most precise method used for evaluation of the patients' clinical outcome. A total of 25% of the patients with localized prostate cancer experience recurrence [1]. Most of the patients with recurrence or metastasis respond to hormone therapy at first, but when hormone therapy fails, the treatment efforts are more limited and the survival rate of the patients will be less than a year. Although the primary results of some chemotherapy medicines are hopeful, we still need some phase-III trials for their confirmation [2].

Progression of the prostate cancer from a normal prostate epithelium to the cancer tissue sensitive or resistant to hormone is a multi-stage process affecting many growth factor signals. Understanding the molecular mechanisms of the prostate cancer helps us find new treatments affecting the genes and their products. Some growth related factors which can determine final prognosis of patients in different tumors are Ki67 expression, EGFR expression and HER-2/neu Oncogene over expression [3-6]. There are some evidences showing a specific role for EGFR gene and the cellular wall receptors [7-9].

The EGFR family genes include c-erB$_1$, c-erB$_2$ (HER$_2$), c-erB$_3$, and c-erB$_4$. They all have an extracellular receptor, to which the legends are attached to form homodimers and heterodimers regarding the activity of intracellular tyrosine kinase which results in cellular proliferation, prevention of apoptosis, and adhesion and cellular attack [10, 11].

Previous studies showed over expression of c-erB$_1$ in primitive and metastatic cancer of the prostate [7, 8]. However, by the time of performing these studies, it was not confirmed if the expression of this receptor was correlated with the pathologic evidences of disease progression [13, 14].

Regarding the biologic differences in prostate cancer and the diversity of the factors that technically affect these results, we decided to evaluate the level of EGFR gene expression in Iran

and show its relation with clinical and pathological evidences of the cancer progression (PSA and grade).

Materials and Methods

The patients with prostate cancer who had referred to Shohada Hospital between 1995 and 2004, had undergone radical prostatectomy, and their tissue samples were confirmed to be adenocarcinoma were enrolled in the study. Two-micron to 3-micron cuts were taken and evaluated for EGFR gene expression by immunohistochemistry method by a pathologist blind to the level of malignancy in the samples. A manual avidine-biotin-peroxidase complex procedure was used in the IHC analysis (DakoCytomation, Copenhagen, Denmark). For this purpose the sections were deparaffinized and processed as follow:

1) The samples were placed in oven at 50-60 C for30 minutes, **2)** The samples were rinsed in 100%Xylol,100%,85%and75% ethanol and water, **3)** Rinsed in 10%PBS, **4)** Rinsed 10 minutes in 1:9 H2O2/ethanol solution, **5)** Rinsed in 10%PBS, **6)** Placed in sodium nitrate buffer(PH=8)and autoclave it for 10 min in 120c and pressure of 1.2 atmosphere, **7)** Rinsed in 10%PBS, **8)** Add 2 drops of serum blocking solution and wait for 10 min, **9)** Cover the slides by 2 drops of primary Ab(her2/neuAb) for 30-60min, **10)** Rinsed in PBS10%, **11)** Add 2 drops of biotinylated Ab on slides and wait for 10min, 12) Rinsed in PBS, **12)** two drops of enzyme conjugate for 10min, 13) Rinsed in PBS, **14)** Add 100 lambda ready DAB chromogen and wait 50min, **15)** Rinsed in PBS, **16)** Two drop of hematoxylin added for contrast making,1-3min and rinsed in water, **17)** PBS 30seconds, **18)** Dehydrated in 75%,85%and 100% alcohols and then 100%xylol for clearing, **19)** Cover slipped and coded.

The immunologic reaction of EGFR was graded as follows: Zero, without reaction; 1, less than 5% of the cells had reaction; 2, between 5% and 50% of the cells had reaction; and 3, more than 50% of the cells had reaction. The tissue factors were considered positive if the grading was 2 to 3[7, 8]. In order to evaluate the relation between clinicopathologic characteristics of EGFR expression, chi-square test was used. A P-value less than .05 was considered statistically significant.

Results

Mean age of the patients was 68 years (range, 45 to 79 years). Of 100 samples evaluated, 80 and 20 had Gleason score ≥ 7 and < 7, respectively. Also, 68 and 32 had PSA level ≤ 10 and >10, respectively. A total of 59 samples were positive for EGFR expression, of whom, 46 had GS < 7 and 13 had GS > 7 (P-value =0 .39). Forty patients had PSA < 10 and 19 had PSA > 10 (P-value = 0.5). The relation between EGFR expression and PSA is (P-value=0.56) and grade is (P-value=0.36).Thus there isn't any significant correlation.

Discussion

According to our results, 59% of the patients were EGFR-positive, which is in accordance with the results of Di Lorenzo and colleagues and Shuch and associates [12, 15]. The expression of this gene is more frequent in black patients suggesting that some genetic factors may affect its expression.

The expression of EGFR gene is quite normal in normal prostate tissue and is dominantly positive in the basal epithelial compartment. The tissue staining is predominantly membranous which is different from tumoral tissues (membranous or cytoplasmic staining) [16].These results show the importance of the regulating role of EGFR for having a normal prostate function. The need for more studies in downstream pairing of EGFR to cellular response in normal tissue in comparison with tumoral tissue is more sensed [17].

We also concluded that the increased expression of this gene is not significantly related to advanced disease which is in contrast with the results of Di Lorenzo's and Shuch's studies. However, in the study by Shuch and co-workers, a significant relationship was found between the EGFR gene expression and PSA but not with grade [15]. Our results are in accordance with the results of Rajal and associates who did not find a significant relationship between PSA, grade, and stage of the disease and postoperative positive margins with EGFR gene expression [16].

Recurrence has been shown in 67% and 8.3% of the patients with positive and negative EGFR expression after radical prostatectomy [12]. The EGFR gene is the only independent factor with prognostic effect on disease-free survival rate and it has been confirmed in this same paper that in 100% of the patients with hormone-resistant prostate cancer, EGFR gene is positive. That is, in EGFR positive patients, the possibility of converting the prostate cancer to a hormone-resistant cancer is higher [12]. Therefore, our future aim is the evaluation of recurrence and disease-free survival rate in these patients and evaluation of gene expression in tissue samples of the patients with hormone-resistant prostate cancer for completion of these studies.

Table 1. EGFR frequency expression in different PSA level

PSA / EGFR expression	≤10 (%)	>10(%)	Total (%)
Negative	28	13	41
Positive	40	19	59
Total	68	32	100

Table 2. EGFR frequency expression in different Grade Score

Grade / EGFR expression	<7(%)	≥7 (%)	Total (%)
Negative	34	7	41
Positive	46	13	59
Total	80	20	100

Since some specific anti-EGFR factors exist in most of the epithelial cancers, [18] a great role is considered for the concurrent use of hormone and EGFR-targeted medications as an alternative treatment in these patients if it is confirmed that the rate of recurrence is high in EGFR-positive patients, as it has been suggested that ZP183 (an inhibitor of EGFR) inhibits the growth of in vivo hormone-resistant cancer [19]. We may prevent the conversion of prostate cancer to a hormone-resistant cancer using these medications.

Conclusion

In 59% of the patients with localized cancer who have undergone radical prostatectomy, EGFR is positive and no significant relation exists between its expression and grade and PSA.

Despite in this study, we did not find relationship between EGFR over expression and some prognostic factors, we recommend other studies with patients follow up.

Acknowledgment

None

Conflict of Interest

The authors have no conflict of interest in this article.

Authors' Contribution

The design, collection of data, interpretation and writing the manuscript were done by all authors.

References

1. Auclerc G, Antoine EC, Cajfinger F, Brunet-Pommeyrol A, Agazia C, et al. Management of advanced prostate cancer. Oncologist 2000; 5:36-44.

2. Petrylak DP. Chemotherapy for advanced hormone refractory prostate cancer. Urology 1999; 54:30–35.

3. Ware JL. Growth factors and their receptors as determinants in the proliferation and metastasis of human prostate cancer. Cancer Metastasis Rev. 1993; 12:287-301.

4. Jalali Nadoushan MR, Taheri T, Jouian N, Zaeri F. Overexpression of HER-2/neu oncogene and transitional cell carcinoma of bladder. Urol J. 2007; 4(3):151-4.

5. Mofid B, Jalali Nodushan MR, Rakhsha A, Zeinali L, Mirzaei H. Relation between HER-2 gene expression and Gleason score in patients with prostate cancer. Urol J. 2007; 4(2):101-4.

6. Jalali Nadoushan MR, Heidary F, Zaeri F, Ahmadi H. Correlation between Grade in Transitional Cell Carcinoma and Expression of Epidermal Growth Factor Receptor. Iranian J Publ Health 2007; 36(2):47-9.

7. Jalali Nadoushan MR, Neisani E, Karbassi M. Correlation of Ki67-Positivity in Tumoral cells Percentage with Effective Factors on prognosis in Primary Breast Cancer. Res J Biol Sci. 2007; 2(3):326-8.

8. Steiner MS. Role of peptide growth factors in the prostate: a review. Urology. 1993; 42:99-110.

9. Russell PJ, Bennett S, Stricker P. Growth factor involvement in progression of prostate cancer. Clin Chem. 1998; 44:705-23.

10. Barton J, Blackledge G, Wakeling A. Growth factors and their receptors: new targets for prostate cancer therapy. Urology. 2001; 58:114-22.

11. Scher HI, Sarkis A, Reuter V, et al. Changing pattern of expression of the epidermal growth factor receptor and transforming growth factor alpha in the progression of prostatic neoplasms. Clin Cancer Res. 1995; 1:545-50.

12. Di Lorenzo G, Tortora G, D'Armiento FP, De Rosa G, Staibano S, Autorino R, et al. Expression of epidermal growth factor receptor correlates with disease relapse and progression to androgen-independence in human prostate cancer. Clin Cancer Res. 2002; 8:3438-44.

13. Hernes E, Fosså SD, Berner A, Otnes B, Nesland JM. Expression of the epidermal growth factor receptor family in prostate carcinoma before and during androgen-independence. Br J Cancer. 2004; 90:449-54.

14. Bartlett JM, Brawley D, Grigor K, Munro AF, Dunne B, Edwards J. Type I receptor tyrosine kinases are associated with hormone escape in prostate cancer. J Pathol. 2005; 205:522-9.

15. Shuch B, Mikhail M, Satagopan J, Lee P, Yee H, Chang C, et al. Racial disparity of epidermal growth factor receptor expression in prostate cancer. J Clin Oncol. 2004; 22:4725-9.

16. Harper ME, Glynne-Jones E, Goddard L, Mathews P, Nicholson RI. Expression of androgen receptor and growth factors in premalignant lesions of the prostate. J Pathol. 1998; 186:169-77.

17. Shah RB, Ghosh D, Elder JT. Epidermal growth factor receptor (ErbB1) expression in prostate cancer progression: correlation with androgen independence. Prostate. 2006; 66:1437-44.

18. Ciardiello F, Tortora G. A novel approach in the treatment of cancer: targeting the epidermal growth factor receptor. Clin Cancer Res. 2001; 7:2958-70.

19. Sirotnak FM, She Y, Lee F, Scher HI. Studies with CWR22 xenograft models in nude mice suggest that ZD1839 ('Iressa') may have a role in the treatment of both androgen-dependent and androgen-independent human prostate cancer [abstract]. Clin Cancer Res. 2001; 7:542.