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Prognostic role of epidermal growth factor receptor in localized breast cancer: 15 years of follow-up

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ABSTRACT

Background: *The expression of epidermal growth factor receptor (EGF-R) in breast cancer (BC) is generally considered as an unfavorable event during tumor progression. Its predictive role has been fairly well defined: EGF-R expression predicts tamoxifen un-responsiveness. EGF-R role in autocrine growth regulation was confirmed. However, reported results on its prognostic role in BC patients were conflicting. The prognostic role of EGF-R after 15 years of follow-up is analyzed in a group of 70 localized BC patients, presented at diagnosis in clinical stages I-III.*

Methods: *BC patients newly diagnosed from December 1990 until March 1991, treated in accordance to the National Protocol, were selected for EGF-R analysis. Steroid receptors and EGF-R were determined at diagnosis in same frozen tissue samples, using biochemical methods. Except 6 patients who were lost from follow-up in the interval shorter than 60 months, the remaining patients were followed-up from 60-188 months. The total number of events was 32 relapses (46%), and 27 deaths (38.5%).*

Results: *EGF-R expression was found in 28/70 patients (40%), and the EGF-R content higher than 26 fmol - in 15/70 patients (21%). Neither the expression, nor the high content of EGF-R showed any infuence on disease-free or overall survival. Levels of EGF-R were similar in relapsing and relapse-free patients. Nodal status had the strongest influence on prognosis.*

Conclusion: *Our results suggest that the controversial findings, regarding the EGF-R prognostic role, might be the consequence of a genuine weak influence of EGF-R expression on disease outcome.*

KEY WORDS: *Breast Neoplasms; Prognosis; Receptors, Steroid; Receptor, Epidermal Growth Factor*

INTRODUCTION

Epidermal growth factor receptor (EGF-R), a member of large type I receptor family, is a membrane protein, which binds ligands, such as EGF or transforming growth factor-α (TGF-α), with the extra cellular region, containing ligand binding domain. This binding leads to the formation of monodimers and heterodimers mostly with cErbB2 (HER2), activating intrinsic tyrosine kinase region of the receptor, which leads to the stimulation of signaling pathways, including the downstream kinases, and triggers the gene transcription, cell proliferation and survival. Various growth factor signal pathways were found to be interactive (1). The so called cross-talk between estrogen receptor (ER) and growth factor (GF) signaling pathways was described, which can promote the escape from the growth inhibition by hormonal treatment, and consecutive resistance to the treatment. Tamoxifen resistant cells may retain ER and remain sensitive to estrogen deprivation with either ovarian ablation (2) or aromatase inhibitors (3). Increased components of EGF-R/HER2 signaling pathways, such as over-stimulation with ligands, increased receptor expression/amplification, increased heterodimerization, or increased downstream pathway elements, were found in acquired and *de novo* tamoxifen resistant BC cells (4), and cell lines (5). Increased GF signaling may also reduce the ER expression (6), which may lead to the insensitivity to all endocrine manipulations (7).

The expression of EGF-R in breast cancer is generally assumed to represent the unfavorable event during the process called "tumor progression". On clinical material, its predictive role has been fairly well defined. Patients whose tumors expressed the EGF-R responded less frequently to endocrine treatment, as compared to those tumors that did not express it (8,9). In these studies, as well as in our own study (10), levels of EGF-R were significantly higher in non-responders, than in responders to endocrine treatment. Interestingly, we found that the EGF-R values mostly were overlapped between responders and non-responders, but did never exceed 26 fmol/mg in those tumors that had regressed on endocrine therapy. Later, it was reported that the EGF-R (and/or HER2) expressing tumors seemed to be less responsive to tamoxifen, but not to aromatase inhibitors (11), which suggested the role of EGF-R in clinical resistance to tamoxifen.

EGF-R is also assumed as a marker of autocrine regulation of tumor growth. TGF-α is one of its ligands, which could be expressed by same tumor cells, forming the autocrine loop. It was found that co-expression of EGF-R and TGF-α in early BC had the most significant effect on Disease-free interval (DFI) and overall survival (OS), higher than nodal status (12).

In accordance with the EGF-R role in endocrine- (tamoxifen-) resistance and autocrine tumor growth regulation, it was expected that the prognosis would be much poorer in patients with EGF-R expressing tumors. However, early results on its prognostic role in breast cancer remained conflicting. Harris and co-workers (13) found that the EGF-R-expressing operable breast cancer patients had shorter DFI and OS. EGF-R-positive tumors progressed more rapidly than EGF-R-negative ones, under the primary endocrine treatment (8). This suggested

that the EGF-R expression was a sign of more aggressive tumor behavior, or in other words, the unfavorable event. However, some authors found the prognostic value only in ER-negative (14-17), or only in node-negative operable breast carcinoma (15,16,18), while others did not find any prognostic significance of EGF-R (19-22). The reason for such controversial findings might be the use of too small patients' groups, too short follow-up, retrospective analysis of the patients' material, and the use of different methods for the EGF-R determination. On the contrary, the genuine weak influence of EGF-R expression on disease outcome could attribute to the inconsistency of the findings. Moreover, within studies that used the biochemical method for EGF-R determination, the lack of the commonly accepted cut-off value for EGF-R positivity could be the major cause of confusion.

In 1996, in a sample of 106 unselected BC patients, we found that the EGF-R binding in individual breast carcinomas was independent of ER/PR, classic prognostic factors (nodal status, tumor size and grade, age and menopausal status) and clinical stage of the disease. It has been concluded that the differences in EGF-R quantitative content represented the true biological difference between tumors (23). This was in contrast to other findings, but not all: some authors found the correlation between EGF-R expression and nodal status (24), or histology grade and even menopausal status (25), or age (26), but others mostly did not find such correlation. In the analysis of OS in metastatic disease, we found a trend towards longer survival in patients whose EGF-R content was low, compared to those with the high EGF-R content (27). In the whole group of 106 unselected BC patients, when gradually increased the cut-off value by 5 fmol/mg, from 0 to up to 50 fmol/mg, the prognostic significance became borderline at the level of 20 fmol/mg, and significant at the level of 25, and especially 26 fmol/mg (28). These published data referred to the group of patients unselected according to clinical stage and to the follow-up of 5-years. However, neither low nor high cut-off values of EGF-R had any significant prognostic influence in a subgroup of operable BC patients. Some more recently published studies with large number of patients showed significant influence of EGF-R on both DFI and OS (29,30), while others did not show any prognostic value of EGF-R (26).

In the present paper, the prognostic role of EGF-R content is re-analyzed after 15 years of follow-up, in a subgroup of localized BC patients.

PATIENTS AND METHODS

Patients

Seventy patients with localized breast cancer have been included in present analysis, as the part of a larger sample of unselected consecutive breast cancer patients, newly diagnosed between December 1990 and March 1991 in the Institute of Oncology and Radiology of Serbia, Belgrade (27).

The group consists of operable stage I-II BC patients (N=58), having been subjected to surgery as the primary treatment, and stage III patients (N=12), having been subjected to delayed radical mastectomy with axillary dissection, after primary (preoperative) radiotherapy. They were presented at diagnosis as borderline operable, but became operable due to the response to radiotherapy. Details of patients' characteristics are presented in Table 1.

Patients were considered as premenopausal if they had regular menstrual cycles at the time of diagnosis; postmenopausal - if the last regular cycle occurred 5 or more years earlier; and perimenopause was defined as the interval between pre- and postmenopause.

Treatment

The choice of primary and consecutive treatment was done in accordance to the National Breast Cancer Protocol. The preoperative and/or adjuvant treatment depended on clinical and pathological stage, tumor size, nodal involvement, histological grade, menstrual status and steroid receptor status: patients in early clinical stages (I-II) were routinely treated by surgery (modified radical mastectomy with axillary dissection in 53, and quadrantectomy without axillary dissection in 5 patients). Node-negative patients with histology grade I-II tumors were followed-up without any adjuvant treatment. The adjuvant treatment of high-risk node-negative (N-) patients, defined with tumor grade III, consisted of CMF chemotherapy (cyclophosphamide, methotrexate, 5-fluorouracil) in PR-negative (PR-), and endocrine therapy in PR-positive (PR+) patients (castration by irradiation in premenopausal, tamoxifen in postmenopausal). Node-positive (N+) operable breast cancer patients were treated with adjuvant CMF chemotherapy if they were PR-, and with CMF chemotherapy + endocrine therapy, if PR+. All postmenopausal N+ patients have been irradiated postoperatively. Those patients who were clinically classified at stage IIIa (T1-3, N2) and IIIb (T4b, N1-2) according to TNM classification, were subjected to biopsy only and irradiated preoperatively with tumor dose (TD) 45 Gy to the whole breast, and 45 Gy to regional lymphatics, in 15 fractions both, every second day during 6 weeks. Two months later (if responded), they were subjected to radical mastectomy with axillary dissection. All stage III patients, presented here, responded to radiotherapy. The further adjuvant treatment depended on pre-irradiation tumor status, postoperative patho-histological nodal status, as well as on histological tumor grade and receptor status of primary tumor, on the same way as in patients primarily operated. EGF-R findings were not known at the time of adjuvant treatment making decision.

Table 1. Patients' and tumors' characteristics

Characteristics		No pts.	%
Histologically confirmed invasive BC		70	100
Age	<45	17	24
	45-59	35	50
	>=60	18	26
Menopausal status	Premenopausal	31	44
	Perimenopausal	8	11
	Postmenopausal	31	44
Initial clinical stage	I	18	26
	IIa	33	47
	IIb	7	10
	III	12	17
Tumor size	pT1	36	52
	pT2	26	37
	pT3	3	4
	T4b	5	7
Nodal status	0	32	46
	1-3	12	17
	>=4	8	11
	unknown	18	26
Histology tumor type	IDC	39	56
	ILC	18	26
	Other	11	16
	unknown	2	3
Histology tumor grade	I	8	11.5
	II	51	73
	III	10	14
	unknown	1	1.5
Steroid receptors	ER+	40	57
	PR+	40	57
	ER+ and/or PR+	50	71

Abbreviations: IDC Invasive ductal carcinoma, ILC Invasive lobular carcinoma

The diagnosis of the first relapse has been routinely confirmed using X-ray, ultrasound, radionuclide and clinical examinations, as well as computed tomography (CT) and nuclear magnetic resonance (NMR) evaluation, when necessary.

The overall survival analysis was done after 15 years of follow-up. It was accepted that the breast cancer was the cause of death only in case when the patient had well documented progressive metastatic disease 1-3 months prior to death.

Receptor determination

Steroid receptors (SR) were determined at diagnosis, in tissue samples of breast cancer, using biochemical methods, which was previously described. An EGF-R content was determined in a membrane fraction of the same frozen tissue samples. Specific binding was calculated by subtracting non-specific from total binding. Data were calculated according to Scatchard analysis (31-34).

Statistics

In the analyses of parametric values, Chi-square and Fisher's exact test were used. For comparison of the non-parametric values Man-Whitney-U test was used. Survival functions were presented by Kaplan-Meier method, and comparison of the groups was done by Log rank and Wilcoxon test.

RESULTS

In a whole group, 5 and 6 patients were lost from follow-up in the interval shorter than 60 months (median 54 and 55 months) for relapse and survival, respectively. Remaining patients were followed-up from 60 to 188 months (median 111.5 months for relapse and 128 months for survival, respectively).

The total number of events was 32 relapses (46%), and 27 deaths (38.5%). Only one patient died in the 42nd month of FU from the cause other than breast cancer. She died from the recurrent multiform glioblastoma, without signs of BC recurrence. Other deaths were caused by metastatic BC.

EGF-R values varied from 0-330 (median 0) fmol/mg. Any measurable EGF-R expression (> 0 fmol/mg) was found in 28/70 patients (40%), and the EGF-R content higher than 26 fmol/mg - in 15/70 patients (21%). There was no correlation with either other prognostic factors, or age and menopausal status. However, the significant inverse relationship with both ER and PR was found.

We compared the DFS and OS in two groups of patients - those without the EGF-R expression in their tumors and the group with any measurable EGF-R expression (42 and 28 patients, respectively). There was no difference either in DFS or in OS between groups (Figure 1). When the higher cut-off level was used (≥26 fmol/mg), again there was no difference either in the occurrence of relapse or survival (Figure 2).

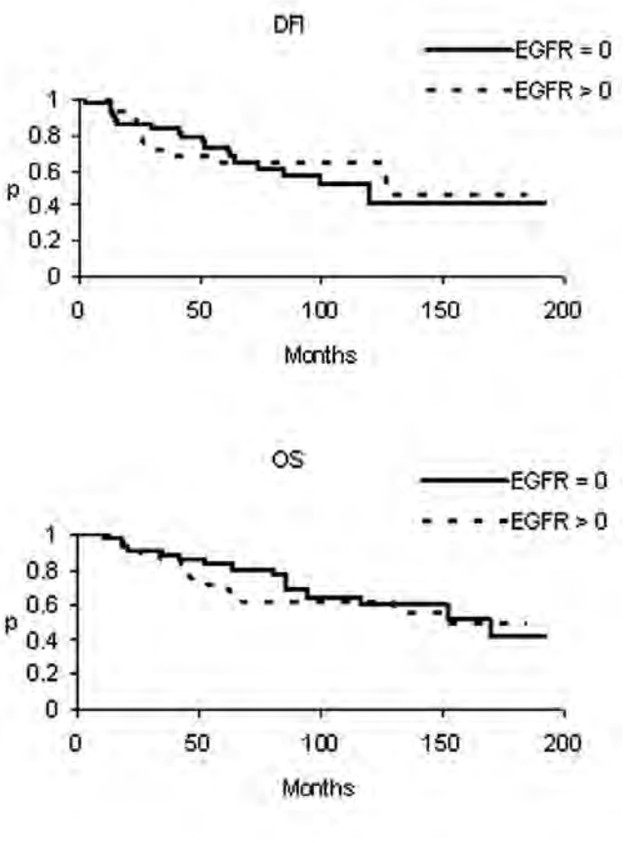

Figure 1. Disease-free interval (DFS) and Overall Survival (OS) in BC patients without the EGF-R expression (EGF-R=0) and any measurable EGF-R (>0 fmol/mg)

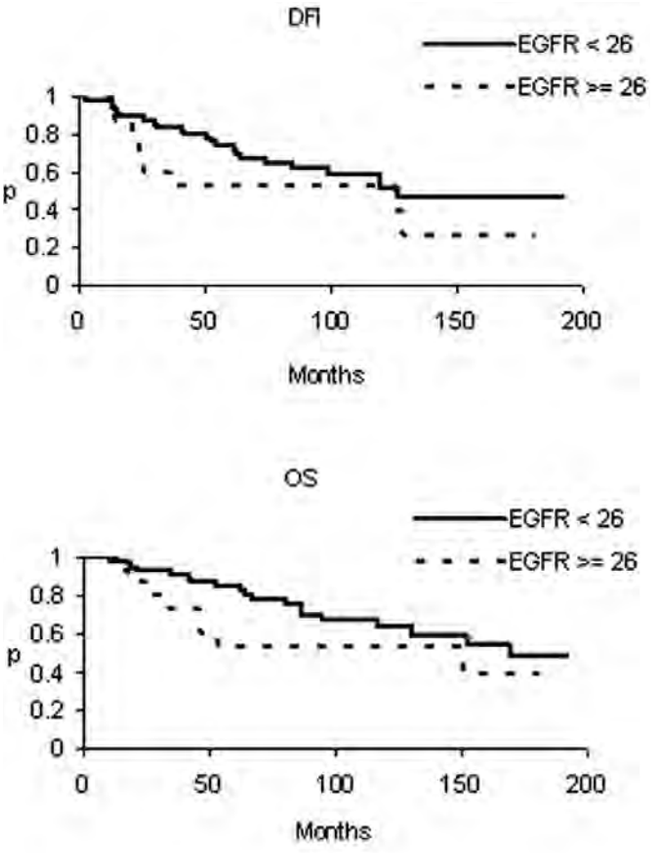

Figure 2. Disease-free interval (DFS) and overall survival (OS) in two subgroups of BC patients in regard to the EGF-R content using the cut-off value of 26 fmol/mg

No difference in the outcome between ER-positive vs. ER-negative patients was found, as well as between PR-positive vs. PR-negative (30 vs. 40 pts both, respectively) (not presented). Moreover, there was no difference in EGF-R content between patients who relapsed and those who remained disease-free (Figure 3).

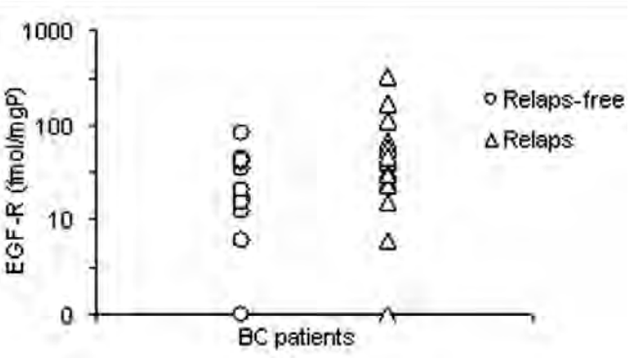

Figure 3. EGF-R content in relapsing and disease free patients

Among other prognostic parameters, nodal status (Figure 4) and clinical stage at diagnosis had the strongest influence on prognosis. All patients were divided by the involvement and the number of involved nodes in pN0, pN1-3, pN≥4, and those with clinical N2 category at diagnosis. Interestingly, both relapse-free and overall survival were similar in node-negative patients and those with 1-3 nodes, and significantly better than in both groups with advanced nodal involvement (N≥4 and those with clinically N2 nodal status). Similar result was found in regards to the clinical stage at diagnosis, concerning the DFI, but not the OS: Stage I and IIa patients had significantly better DFS, than stage IIb and III patients (not presented).

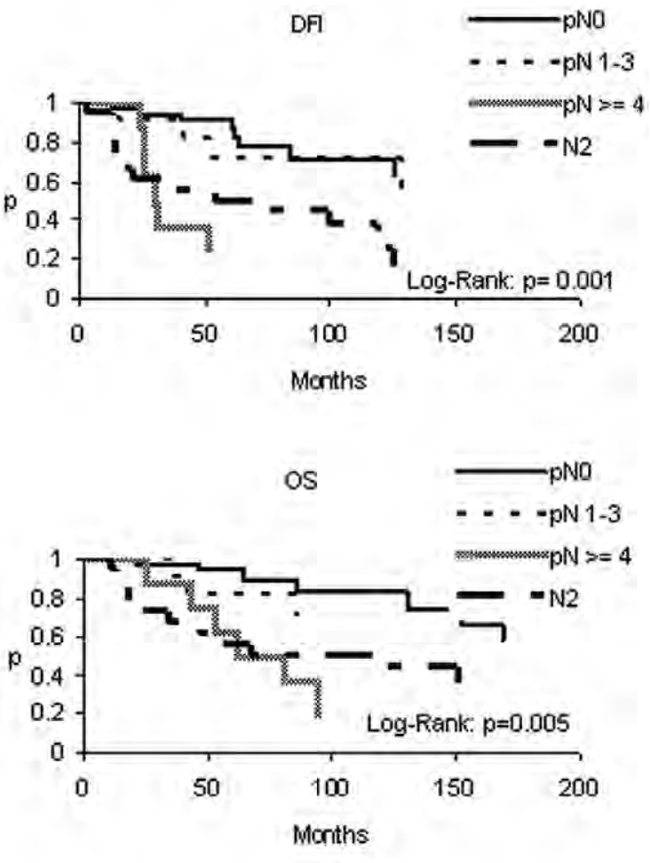

Figure 4. Disease-free interval (DFS) and overall survival (OS) with respect to the nodal status (N) of BC patients

DISSCUSSION

Although the expression of growth factor receptors is commonly thought to be the unfavorable prognostic event, our results, published earlier, showed that a prognostic role of EGF-R expression in early breast cancer patients was negligible (27). This was in accordance with the other findings of a weak, if any, influence of EGF-R on the outcome of operable breast cancer (16,26).

After the follow-up of 15 years, neither the expression, nor the higher cut-offs, when used in a group of operable breast cancer patients, did not change our previous results: there was no difference in the outcome between EGF-R negative and EGF-R positive localized BC patients. In addition, the EGF-R content, determined in relapsing patients, in comparison to those who were still relapse-free, showed no significant difference.

The inverse relationship between EGF-R and ER, often reported in clinical studies (35) was also confirmed in this study: ER+ tumors more frequently had no EGF-R, or its content was low, in comparison with ER- tumors, but the direct correlation of their levels was not confirmed, as reported earlier (23). The commonly used term "inverse" relationship obviously refers to the proportion of ER positive and EGF-R positive cells within a whole tumor. Therefore, it does not exclude the independence of their expression.

In our previous analysis, we found that patients, whose EGF-R content was below 26 fmol/mg, had significantly better prognosis than those with EGF-R content above this cut-off level (28). It seemed that the tumors with EGF-R content lower than this cut-off could be estrogen dependent (10). These results were obtained on the larger unselected BC patients` group, but in the group of operable BC patients, at the point of 5 years follow-up, there was only a non-significant trend towards the influence of EGF-R on outcome. Similar results were published by Foekens (36) and Klijn (37) who found, in operable breast cancer patients, that the group with "intermediate" content (0.5-2.0 fmol/mg) had lower risk of recurrence than both groups with low EGF-R content (<0.5 fmol/mg) and those with high EGF-R content. The offered explanation for this phenomenon implies the possibility that tumors with low EGF-R content do not secrete TGF-α, they do not display autocrine-regulated growth, and consequently they may be endocrine-responsive. The alternative explanation suggested that receptors for EGF could be saturated and masked by their ligand (38).

We have previously found that certain carcinomas were still endocrine regulated and endocrine sensitive as well, although contained EGF-R (10). Nicholson et al (39) found different levels of endocrine insensitivity between highly and moderately EGF-R-positive tumors, according to the level of immunostaining. Therefore, it could be concluded that the quantitative EGF-R content is associated with the level of the loss of endocrine sensitivity. In addition, Nicholson et al (8,9) found the lower levels of EGF-R in responders, than in non-responders to endocrine therapy. In our study, all patients with endocrine responsive tumors received adjuvant tamoxifen or ovarian ablation: their outcome must be changed (improved) in regards with the "natural" course of disease. Thus, it seems noteworthy to analyze the prognostic value of EGF-R in the subgroup of patients who did not receive any adjuvant treatment.

Obviously the expression of EGF-R, which is related to the increased EGF-R/HER2 signaling, leading to increased cell proliferation and survival, tamoxifen and probably endocrine insensitivity and much aggressive tumor behavior – represents the unfavorable event. However, our results, presented here, show that the weak prognostic influence of EGF-R level disappeared after 15 years of FU. Similar loss of prognostic influence was confirmed by Klijn et al. (37).

It is unusual that expression of EGF-R does not influence directly the outcome of early BC patients. Controversial reports still appear (40). Even in studies that confirm the prognostic value of EGF-R (30), the real clinical usefulness of this finding is questionable. EGF-R probably is one of the most important factors in the complex orchestra of growth regulation and endocrine sensitivity/resistance mechanisms. Thus, the further investigation of co-expression and co-operation of EGF-R with other factors, such as HER2, TGF-α and others, could probably be of clinical interest, as well as the investigation of the role of elevated multiple elements of signaling pathways in BC prognosis (41).

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REFERENCES

1. Nicholson RI, Hutcheson IR, Knowlden JM, Jones HE, Harper ME, Jordan N, et al. Nonendocrine pathways and endocrine resistance: observations with antiestrogens and signal transduction inhibitors in combination. Clin Canc Res 2004;10:346S-54S.
2. Love RR, Duc NB, Havighurst TC, Mohsin SK, Yhang Q, DeMets DL, Allread DC. Her-2/neu overexpresion and response to oophorectomy plus tamoxifen adjuvant therapy in estrogen receptor-positive premenopausal women with operable breast cancer. J Clin Oncol 2003;21(3):453-7.
3. Show J, Massarweh S, Osborn CK, Wakeling AE, Ali S, Weiss H, Schiff R. Mechanisms of tamoxifen resistance: increased estrogen receptor-HER2/neu cross-talk in ER/HER2-positive breast cancer. J Natl Cancer Inst 2004;96:926-35.
4. Lipton A, Leitzel K, Ali SM, Demers L, Harvey HA, Chaudri-Ross HA, et al. Serum Her-2/neu conversion to positive at the time of cancer progression in metastatic breast patients treated with letrozole vs. tamoxifen. Proc Am Soc Clin Oncol 2003,22:3 (Abstract 8).
5. Gee JM, Robertson JF, Gutteridge E, Ellis IO, Pinder SE, Rubini M, Nicholson RI. Epidermal growth factor receptor/HER2/insulin-like growth factor receptor activity in clinical breast cancer. Endocrine-Rel Cancer 2005;12:S99-S111.
6. Stoica A, Saceda M, Doraiswamy VL, Coleman C, Martin MB. Regulation of estrogen receptor-alpha gene expression by epidermal growth factor. J Endocrinol 2000;165:371-8.
7. Nicholson RI, Staka C, Boyns F, Hutcheson IR Gee JM. Growth factor-driven mechanisms associated with resistance to estrogen deprivation in breast cancer: new opportunities for therapy. Endocrine-Rel Cancer 2004;11:623-41.

8. Nicholson S, Halcrow P, Sainsbury JRC, Angus B, Chambers P, Farndon JR, et al. Epidermal growth factor receptor (EGFr) status associated with failure of primary endocrine therapy in elderly postmenopausal patients with breast cancer. Br J Cancer 1988;58:810-4.
9. Nicholson S, Sainsbury C, Halcrow P, Chambers P, Farndon JR, Harris AL. Expression of epidermal growth factor receptors associated with lack of response to endocrine therapy in recurrent breast cancer. Lancet 1989;1:182-5
10. Neskovic-Konstantinovic ZB, Nikolic-Vukosavljevic DB, Brankovic-Magic MV, Mitrovic LB, Spuzic I. Content of epidermal growth factor receptor in metastatic breast cancer: its role in endocrine sensitivity prediction. Neoplasma 2000;47(2):107-13.
11. Ellis MJ, Coop A, Singh B, Mauriac L, Llombert-Cussac A, Janicke F, et al. Letrozole is more effective neoadjuvant endocrine therapy than tamoxifen for erbB-1- and/or erbB-2-positive, estrogen receptor-positive primary breast cancer: Evidence from a phase III randomized trial. J Clin Oncol 2001;19:3808–16.
12. Umekita Y, Ohi Y, Sagara Y, Yoshida H. Co-expression of epidermal growth factor receptor and transforming growth factor-alpha predicts worse prognosis in breast-cancer patients. Int J Cancer 2000;89(6):484-7.
13. Harris AL, Nicholson S, Sainsbury JRC, Farndon J, Wright C. Epidermal growth factor receptors in breast cancer: association with early relapse and death, poor response to hormones and interactions with neu. J Steroid Biochem 1989;34:123-31.
14. Nicholson S, Wright C, Sainsbury JRC, Halcrow P, Kelly P, Angus B, et al. Epidermal growth factor receptor (EGFr) as a marker for poor prognosis in node-negative breast cancer patients: Neu and tamoxifen failure. J Steroid Biochem Mol Biol 1990;37:811-4.
15. Railo M, Smitten KV, Pekonen F. The prognostic value of epidermal growth factor receptor (EGFR) in breast cancer patients. Results of a follow-up study on 149 patients. Acta Oncologica 1994;33:13-7.
16. Sainsbury JRC, Needham GK, Farndon JR, Malcolm AJ, Harris AL. Epidermal-growth-factor receptor status as predictor of early recurrence of and death from breast cancer. Lancet 1987;1:1398-402.
17. Lewis S, Locker A, Todd JH, Bell JA, Nicholson R, Elston CW, et al. Expression of epidermal growth factor receptor in breast carcinoma. J Clin Pathol 1990;43:385-9.
18. Fox SB, Smith K, Hollyer J, Greenall M, Hastrich D, Harris AL. The epidermal growth factor receptor as a prognostic marker: Results of 370 patients and review of 3009 patients. Breast Cancer Res Treat 1994;29:41-9.
19. Grimaux M, Romain S, Remvikos Y, Martin PM, Magdelenat H. Prognostic value of epidermal growth factor receptor in node-positive breast cancer. Breast Cancer Res Treat 1989;14:77-99.
20. Toi M, Nakamura T, Mukaida H, Wada T, Osaki A, Yamada H, et al. Relationship between epidermal growth factor receptor status and various prognostic factors in human breast cancer. Cancer 1990;65:1980-4.
21. Gohring U-J, Ahr A, Scharl A, Weisner V, Neuhaus W,Grombach G, et al.. Immunocytochemical detection of epidermal growth factor receptor lacks prognostic significance for breast carcinoma. J Soc Gynecol Invest 1995;2:653-9.
22. Pirinen R, Lipponen P, Syrjanen K. Expression of epidermal growth factor receptor (EGFR) in breast cancer as related to clinical, prognostic and cytometric factors. Anticancer Res 1995;15:2835-40.
23. Neskovic-Konstantinovic Z, Nikolic-Vukosavljevic D, Brankovic-Magic M, Kanjer K, Spuzic I. The content of epidermal growth factor receptor in breast cancer: Association with classical parameters of prognosis and steroid receptor status. J BUON 1996;1:53-7.
24. Ray A, Naik SL, Sharma BK. Distribution of prognostically unfavorable product of c-erbB-2 oncogene and EGF-r in carcinomas of the breast and uterine cervix. Indian J Physiol Pharmacol 2002;46(4):423-33.
25. Ray A, Sharma BK, Kaur S, Sharma S, Sharma JK. Overexpression of c-erbB-2 oncoprotein and associated pathobiological factors in invasive primary breast cancer. Indian J Exp Biol 2004;42(3):253-8.
26. Ferrero JM, Ramaioli A, Largillier R, Formento JL, Francoual M, Ettore F, et al. Epidermal growth factor receptor expression in 780 breast cancer patients: A reappraisal of the prognostic value based on an eight-year median follow-up. Ann Oncol 2001;12:841-6.
27. Neskovic-Konstantinovic Z, Nikolic -Vukosavljevic D, Brankovic-Magic M, Kanjer K, Gavrilovic D, Mitrovic L, et al. Expression of epidermal growth factor receptor in breast cancer, from early stages to advanced disease. J Exp Clin Cancer Res 1999;18(3):347-55.
28. Nešković-Konstantinović Z, Nikolić-Vukosavljević D, Kanjer K, Gavrilović D, Spuzić I. EGF-R as a prognostic biomarker of breast carcinoma: cut-off value. J Tumor Marker Oncol 2004;19(3):187-97.
29. Buchholz TA, Tu X, Ang KK, Esteva FJ, Kuerer HM, Pusztai L, et al. Epidermal growth factor receptor expression correlates with poor survival in patients who have breast carcinoma treated with doxorubicine-based neoadjuvant chemotherapy. Cancer 2005;104(4):676-81.
30. Tsutui S, Ohno S, Murakami S, Hachitanida Y, Oda S. Prognostic value of epidermal growth factor receptor (EGFR) and its relationship to the estrogen receptor status in 1029 patients with breast cancer. Breast Cancer Res Treatment 2002;71(1):67-75.
31. EORTC Breast Cancer Cooperative Group. Revision of the standards for the assessment of hormone receptors on human breast cancer. Eur J Cancer 1980;16:1513-5.
32. Wittlif JL, Pasio R, Bland KI. Steroid and peptide hormone receptor identified in breast tissue. In: Bland KI, Copeland III EM, editors. The breast - Comprehensive management of benign and malignant disease. Philadelphia: Sounders; 1991. p. 900-36.
33. Nikolic-Vukosavljevic D, Kanjer K, Brankovic-Magic M, Spuzic I. Association among an autocrine parameter (EGF-R) and endocrine parameters (ER and PR) in locoregional breast cancer. Neoplasma 1994;41:137-40.
34. Nikolic-Vukosavljevic D, Brankovic-Magic M, Polic Dj, Jankovic R. Standardization of steroid hormone receptor assay. Jugoslav Med Biochem 1998;17(4):391-4.
35. Konecny G, Pauletti G, Pegram M, Untch M, Dandekar S, Aguilar Z, et al. Quantitative association between HER2/neu and steroid hormone receptors in hormone receptor-positive primary breast cancer. J Natl Cancer Inst 2003; 95(2):142-53.
36. Foekens JA, Portengen H, Van Putten WLJ, Trapman AMAC, Reubi J-C, Alexieva-Figusch J, Klijn JGM. Prognostic value of receptors for insulin-like growth factor 1, somatostatine, and epidermal growth factor in human breast cancer. Cancer Res 1989;49:7002-9.
37. Klijn JGM, Look MP, Portengen H, Alexieva-Figusch J, van Putten WL, Foekens JA. The prognostic value of epidermal growth factor receptor (EGF-R) in primary breast cancer: results of a 10 year follow-up study. Breast Cancer Res Treat 1994;29:73-83.
38. Artagaveytia N, Le Penven S, Falette N, Lucero R, Garofalo EG, Saez S. Epidermal growth factor and transforming growth factor alpha mRNA expression in human breast cancer biopsies; analysis in relation to estradiol, progesterone and EGF receptor content. J Steroid Biochem Mol Biol 1997;60(3-4):221-8.
39. Nicholson RI, McClelland RA, Finlay P, Eaton CL, Gullick WJ, Dixon AR, et al. Relationship between EGF-R, c-erbB-2 protein expression and Ki67 immunostaining in breast cancer and hormone sensitivity. Eur J Cancer 1993;29A:1018-23.
40. Rampaul RS, Pinder SE, Wencyk PM, Nicholson RI, Blamey RW, Robertson JF, et al. Epidermal growth factor receptor status in operable invasive breast cancer: Is it of any prognostic value? Letter to the Editor. Clin Cancer Res 2004;10: 2578.
41. Knowlden JM, Hutcheson IR, Jones HE, Madden T, Gee JMW, Harper ME, et al. Elevated levels of epidermal growth factor receptor/c-erbB2 heterodimers mediate an autocrine growth regulatory pathway in tamoxifen-resistant MCF-7 cells. Endocrinology 2003;144(3):1032-44.