RESEARCH COMMUNICATION

Clinicopathologic Characteristics and Prognostic Factors in Patients with Operable HER-2 Overexpressing Breast Cancer

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Abstract

Objective: To study the relationship between clinical pathologic characteristics, treatment modalities and prognostic factors in HER-2 (Human Epidermal growth factor Receptor-2) overexpressed breast carcinoma. Materials and Methods: Major clinico-pathological factors including therapeutic modalities and survival status of 371 breast cancer patients with HER2 over-expression, teated at Yantai Yuhuangding Hospital from March of 2002 to December of 2010 were retrospectively studied, with special attention focused on survival-related factors. Results: The median age of the total 371 patients in this study was 48 years at time of diagnosis, among which, the leading pathological type was infiltrating ductal carcinoma (92.5%); 62.8% presented with a primary tomor larger than 2 cm in diameter at diagnosis, 51.0% had axillary lymph node (ALN) metastases; ER (Estrogen receptor) /PR (Progesterone receptor) double negative occured in 52.8% of cases, and PCNA (proliferation cell nuclear antigen) (+++) was found in 55.1%. HER-2 overexpressed patients were usually in advanced stage when the diagnosis was made (72.8% at stages IIA~IIIC). The prognosis and survival were assessed in 259 patients with complete follow-up data. 5-year DFS (disease-free survival) and OS (overall survival) rate was 68.0% and 78.0% respectively. Univariate analysis revealed that age, tumor size, ALN metastases, LVSI (lymph-vascular space involvement), PCNA status, hormonal therapy, chemotherapy cycles, and HER-2 overexpression, correlated closely with the prognosis. ALN metastases, LVSI, PCNA status and chemotherapy cycles were independent predictors of survival. Conclusions: HER-2 overexpressed breast cancer has special clinical and pathological characteristics, with advanced clinical stages and high rate of ER/PR double negative. Lymph node metastases, LVSI, PCNA and chemotherapy cycles are independent predictors of prognosis.

Keywords: Breast cancer - HER2 overexpression - clinicopathology - prognosis - multivariate analysis

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Introduction

Breast cancer is one of the most common malignant tumors in women. Recently breast cancer has increased greatly in incidence and is now the leading malignant disease in a few areas such as Shanghai in China (Tang, 2000). HER-2 overexpression occurs in approximately 20-30% breast cancer patients. Such overexpression points to a particular type of malignant tumor with special clinical and histological characteristics. HER-2 overexpression often indicates earlier tumor reccurrence along with a shorter survival duration (Revillion et al., 1998; Ross et al., 1998).

The clinical pathological characteristics of 317 breast cancer patients with HER-2 overexpression treated at our hospital were studied. Of these patients, 259 had complete follow-up data and formed the patient group for studies to elucidate survival-related factors.

Materials and Methods

Clinical data

We reviewed all breast cancer patiens who had undergone a surgery in Yantai Yuhuangding Hospital from March of 2002 to December of 2008. A total of 371 patients had tumors with HER-2 (+++) as identified by immunohistochemical (IHC) tests. Among these cases, 259 had complete treatment data available for analysis; by July of 2011, 246 were followed up by phone call or communicate by letter, 5% were lost; duration of follow-up ranged 5.0~96.0 months, the median was 48.0 months.

Methods

Overall survival (OS) was defined as the time from diagnosis to death for any cause or the last visit. Disease-free survival (DFS) was the time from diagnosis to recurrence/metastasis at any part of the body.

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Software SPSS13.0 was used for statistical analysis. Kaplan-Meier method was used for survival analysis; Log-rank test was adopted for comparison between groups; and COX proportional hazard regression model was selected for use in prognosis analysis.

Results

Clinical pathologic features of the patients

Major clinico-pathological features of 371 patients with available pathological data and HER-2 overexpression (3+, IHC staining) were detailed Tables.

Treatment received by the patients

Among all 371 cases, 259 had complete data of post-operation treatment available for analysis, in whom 228 (88%) received chemotherapy after operation; 185 (71.4%) received 4-6 cycles of chemotherapy, 27<4 cycles, 17>6 cycles; 221 received Anthracyclines-based chemotherapy, only 6 had CMF (cyclophophmide, metroxtrate, 5-fluoreuricil) regimen; adjuvant hormonal therapy was performed in 133 patients (53.3%) after surgery, 33 (12.7%) received Trastuzumab targeted therapy for a duration of 1 year or 2 years after chemo/radio therapy. 5.0~96.0 months, the median was 48.0 months.

Recurrence, metastases and survival

By July of 2011, the median follow-up was 48.0 (range 5.0-96.0) months. Among the 259 patients with available data, 177 (24.7%) remained disease-free, 64 had local recurrence and/or distant metastases, second primary tumor occurred in 5 cases (1.9%), and 13 patients (5.0%) were lost for follow-up.

The incidence of local recurrence (chest wall, lymph node, and skin etc.) after operation was 41.5% (n=107); Viceral metastases (spread to liver, lung, brain etc.) accounted for 44.6% (n=116). Median recurrence/metastasis interval was 23.5 months (range from 2~78 months), suggesting that recurrence peaked within 2 years after operation.

Overall 5-year DFS and OS was 68.0% and 78.0% respectively. (See Figure 1-2) At the end of follow-up, there were total 41 deaths, one from cardio-and cerebro-vascular disease, one from second primary malignancy, all others died of recurrence/metastases of breast cancer. Among the 259 patients with complete data available for analysis, 5 (1.9%) occurred second primary tumor. Median interval between appearance of second primary malignancy and diagnosis of breast cancer was 17.8 months; 3 were contralateral breast cancer, and 1 was endometrial carcinoma.

Univariate analysis of prognostic factors

Clinical pathological factors were analysed by using Univariate Kaplan-Meier survival analysis (Log-rank test). The results revealed that age, tumor size, lymph node metastases, LVSI, hormonal therapy, chemo cycles and PCNA overexpression correlated closely with the outcome (Table 1). 5-year DFS and OS rates decreased significantly in patients associated with any of the following factors: age<35 years; tumor size greater than 2 cm; ALN metastases; LVSI; PCNA (+++); hormonal therapy, P value were all <0.05. Compared with the group in which chemotherapy was used less than 4 cycles, 5-year DFS and OS rates improved markedly in the group with >4 cycles (p<0.001). Factors including hormone-receptor

Table 1 Univariate Analysis for Prognosis in 259 Operable Breast Cancer Patients with HER-2 Overexpression

	5-yr OS (%)	P value	5-yr DFS (%)	P value
Menopausal status				
Pre-menopausal	63.8	0.05	54.0	0.03
Post-menopausal	90.4		73.6	
Histological grade				
Grade 2	82.6	0.08	73.7	0.39
Grade 3	66.6		60.9	
Tumor size				
T≤2cm	90.6	0.02	82.2	0.02
T>2cm	73.7		65.6	
ALN metastases				
Negative	89.8	<0.001	86.4	<0.001
Positive	70.5		58.5	
ALN metastases (number)				
0	89.8	All <0.05	86.4	All <0.05
1-3	77.1		62.2	
4-9	71.6		64.6	
≥10	60.0		38.5	
LVSI				
None	83.6	0.00	75.8	0.01
Exist	64.3		57.1	
ER and PR status				
Double negative	75.8	0.15	72.8	0.59
Positive (ER and/or PR)	83.4		71.9	
P53				
+++	85.5	0.44	68.0	0.68
-~++	86.9		72.7	
PCNA				
+++	71.6	0.00	72.5	0.02
-~++	90.1		78.0	
Chemotherapy				
Y	79.5	0.69	82.1	0.63
N	80.4		70.4	
Chemo regimen				
Non-anthracyclines	66.7	0.48	66.7	0.45
Anthracyclines	78.6		67.1	
Paclitaxel+ Anthracyclines	80.7		77.5	
Chemo cylces				
<4	55.8	<0.001	41.8	<0.001
4-6	83.5		74.4	
>6	81.7		74.7	
Radiotherapy				
Y	77.3	0.35	65.4	0.06
N	80.9		76.2	
Hormonal therapy				
Y	84.3	0.04	77.0	0.03
N	75.3		67.1	
Trastuzumab therapy				
Y	93.8	0.09	84.4	0.09
N	77.6		70.2	
Duration of Trastuzumab therapy				
1 year	95.2	0.66	85.0	0.62
2 years	90.9		85.7	

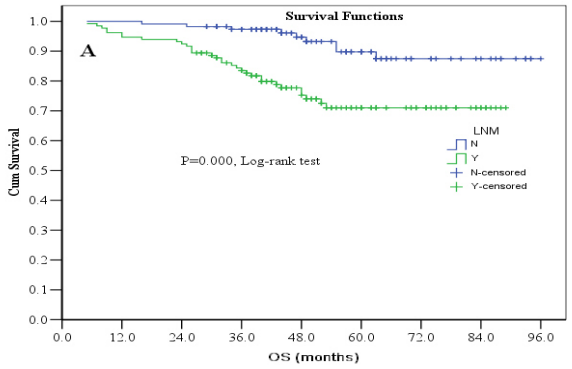

Figure 1. Kaplan-Meier Analysis Showed that Lymph Nodes Metastasis Had a Significant Impact on Both OS (A) and DFS (B). Y: LNM; N: no LNM

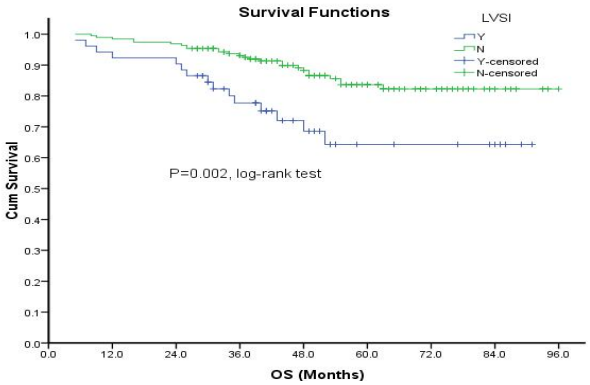

Figure 2 Kaplan-Meier Analysis Showed that LVSI Had a Significant Impact on Both OS (A) and DFS (B)

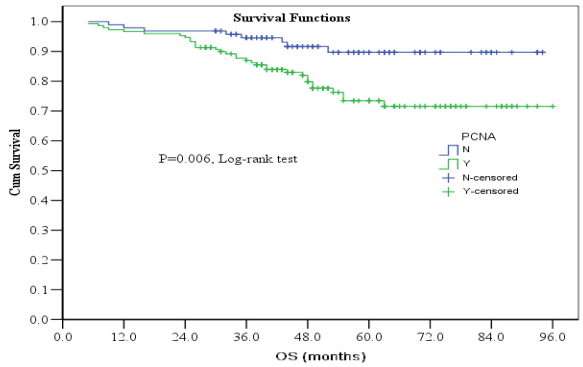

Figure 3 Kaplan-Meier Analysis Showed that PCNA (3+) Had a Significant Impact on Both OS (A) and DFS (B). Y:PCNA>=3+; N: PCNA<3+

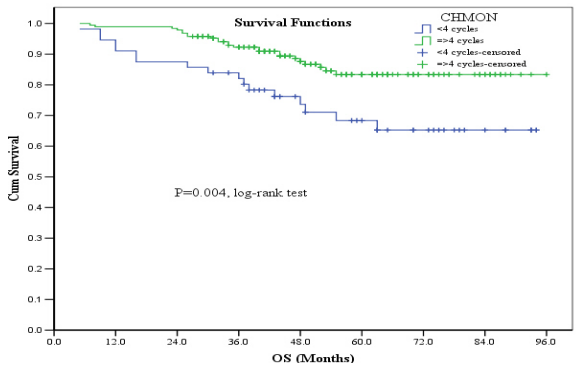

Figure 4 Kaplan-Meier Analysis Showed that Chemotherapy over 4 Cycles Had a Significant Impact on Both OS (A) and DFS (B)

status, P53 expression, whether or not received chemo/radio therapy, and difference of chemotherapy regimen had little effect on survival.

Table 2. COX Multivariate Analysis for Prognosis in 259 Operatable Breast Cancer Patients with HER-2 Overexpression

Prognostic factors	P value	95% confidence interval
LN metastases	0.01	1.35~6.35
LVSI	0.01	1.21~4.66
PCNA (+++)	0.00	1.41~6.66
Chemo≥4 cycles	0.00	0.18~0.64

Multivariate analysis of prognositic factors

COX multivariate regression analysis was performed to further investigate the 7 prognostic factors appeared with statistical significance in the above univariate analysis. The 7 prognostic factors were age, tumor size, lymph node metastases, LVSI, PCNA status, the number of chemotherapy cycles and hormonal therapy. It was revealed that lymph node metastases (Figure 1), LVSI (Figure 2), PCNA status (Figure 3), and chemotherapy cycles (Fiugre 4) were independent predictors of 5-year OS and DFS (Table 2).

Discussion

This study has shown that HER-2 overexprssion breast carcinoma most commonly occurred between 41~60 years old (61.8%), especially before menopause (61.2%). The most common histological type was infilitrating ductal carcinoma (92.5%), with more than one half had a primary tumor greater than 2 cm at diagnosis; most patients were in locally advanced stages when diagnosis was made (72.8%, stagesIIA~IIIC); ER/PR double negative occurred in 52.8% of patients, and PCNA (+++) accounted for 55.1%. This indicates that most patients with HER-2 overexprssion were in locally advanced stages at diagnosis, with active cell proliferation properties, and requires aggressive adjuvant treatment after operation.

A prognosis analysis performed (Ferrero-Pous et al., 2000) in 488 operable breast cancer patients demonstrated that 5-year DFS and OS was 62.6% and 67.7% respectively in HER-2 overexpression group, both differences were significant. Another study (Xia et al., 2004) revealed that 5-year OS was approximately 60% in patients with HER-2 overexprssion breast cancer. In our study, 5-year DFS and OS of 259 patients was 68.0% and 78.0% respectively, quite similar to the published results.

Young age (<35 yr) was proved to be an independent prognostic factor for poor outcome of breast cancer (Yildirim et al., 2000). Our study demonstrated that both OS and DFS substantially shortened in patients <35 years when compared with those aged ≥35, supporting the notion that younger breast cancer patients with HER-2 overexpression had much worse prognosis.

Tumor size is recognized as an independent predictor for poor prognosis of breast cancer, and distant metastases occur more common in patients with a massive lump (Cianfrocca et al., 2004). Another study (Menard et al., 2002) found in 1,928 patients that tumor size (>2 cm) correlated closely with poor outcome of HER-2 overexprssed breast cancer. A third study (Traina et al.,

2006) had the same conclusion from a study in 1,355 patients with early breast carcinoma. Our result was similar: patients with a tumor greater than 2 cm had significant reduction in OS (p=0.015) and DFS (p=0.019).

Lymph nodes metastasis is the most important prognostic factor in patients with operable breast cancer, and the number of positive lymph nodes correlated directly to recurrence/metastases (Nemoto et al., 1983). It was revealed in an NSABP study (Fisher et al., 1983) that 5-year OS was 82.8% in lymph node negative breast cancer, reduced to 73% in patients with 1~3 positve lymph nodes, 45.7% in patients with 4~12 positive nodes, and 28.4% in those with ≥ 13 positive lymph nodes. In our study, in lymph positive patients, when compared with lymph node negative, 5-year OS was reduced by 19.4% (P<0.001) and DFS reduced by 27.9% (P<0.001), similar to documented literature. Further analysis found that, compared with LNM 1-3 and 4-9 subgroups, DFS of LNM≥10 subgroup reduced significantly (38.5% VS 62.2% and 64.6%, P<0.05) , but there was no statistical difference in OS. This indicates that involvement of 10 lymph nodes and more may be another predictor for poor outcome in HER-2 overexprssion breast carcinoma.

A study of 644 breast cancer patients (Rosen et al., 1989) found with median follow up of 18 years that LVSI indicated worse prognosis, with the recurrence rate in patients with and without LVSI was 38% and 22% respectively. In a study of 1,275 patients, the International (Ludwig) Breast Cancer Study Group (Neville et al., 1992) found that whether or not with adjuvant therapy after operation, 5-year recurrence rate increased by 15% in patients with LVSI. A second study (Faneyte et al., 2004) also found that LVSI was a strong predictor for poor prognosis, DFS and OS might be reduced in these patients. However, another study (Traina et al., 2006) did not find an association between LVSI and poor outcome in patients with HER-2 overexpression. In our study, 5-year OS and DFS in patients with and without LVSI was 64.3% vs 83.6% (P<0.001) and 58.5% vs 86.4% (P<0.001) respectively, suggesting LVSI was a prognostic factor for poor outcome. Multivariate analysis also demonstrated that this was an independent predictor for OS (P=0.012).

Proliferating cell nuclear antigen (PCNA) is a biomarker of cell proliferation expressed at stages S and G2 in a cell cycle (Aaltomaa et al., 1993). PCNA strong positive (+++) indicates an active cell growth, and tends to be a prognostic factor as other markers (ER, PR, HER-2) for breast cancer (Sledge et al., 2003). Although the role of PCNA in prognosis of breast cancer remains uncertain, some evidence (Tahan et al., 1993) demonstrating that PCNA tends to be responsible for the significant reduction in OS and DFS. In our study, PCNA correlated closely to OS and DFS in HER-2 overexpression breast carcinoma, and multivariate analysis also revealed it to be an independent predictor of prognosis (P=0.005).

Receptor status is a predictor for the outcome of hormonal therapy, but its effect on survival remains controversial. It was similar in our study to the reported investigation (Traina et al., 2006), receptor status had little influence on OS and DFS (P=0.15, P=0.59).

According to the results of Meta analysis (Le Doussal et al., 1989), CMF adjuvant chemotherapy has been demonstrated to reduce the risk of recurrence and mortality rates by 24% and 13% respectively; CMF regimen+Anthracyclines might further reduce recurrenc and death rates by 12% and 11%. HER-2 overexpression breast carcinoma has been proven (1998) to be resistant to CMF, while still relatively sensitive to anthracycline-based regimen. In our study, there was no significant difference in DFS and OS between patients with and without post-operative adjuvant chemotherapy, which might be associated with the fact that most patients received chemotherapy (230 cases, 88.8%) and only a few without (29 cases, 11.2%). No significant difference was found in DFS and OS between each different chemotherapy regimen in our study, possibly because of the fact that anthracycline-based regimen accounted for the vast majority, non-anthracycline-containing regimen (CMF) was performed in merely 6 patients (2.3%). In addition, univariate analysis of our study has revealed that chemotherapy less than 4 cycles is another prognostic factor of poor outcome for patients with operable HER-2 overexprssion breast cancer.

It has demonstrated a system review that post-operative adjuvant TAM therapy could significantly improve survival, reduce the risk of recurrence and contralateral breast involvement of breast cancer (Moliterni et al., 2003). It has also been proven (Lancet.,1998; Ellis et al., 2001) that HER-2 overexpression tumor may be unresponsive or resistant to TAM regimen in spite of receptor positive property. In our study, total 133 patients (51.4%) received hormonal therapy, among which 92.5% accepted TAM. The use of hormonal therapy was superior to no endocrine therapy in DFS (77.0% vs 67.1%, P=0.029) and OS (84.3% vs 75.3%, P=0.037) , suggesting that TAM should not be lightly given up in patients with HER-2 overexprssion. In stratified analysis of this study, among the ER and /or PR positive patients, hormone therapy has induced a significant improvement in DFS and OS (P<0.001); Whereas in those appeared ER/PR double negative, endocrine therapy failed to improve DFS and OS.

The role of Trastuzumab as an adjuvant therapy for breast cancer has been initially established by a few large-scale clinical trials (Piccart-Gebhart et al., 2005; Romond et al., 2005; Slamon et al., 2005). Trastuzumab has been proven to potentiate the effect of chemo/radio therapy to further reduce the risk of 1-and 2-year recurrence by 39% to 52%. In addition, a recent study (Gianni et al., 2011) found at a 4-year follow-up that post-operative adjuvant treatment with Trastuzumab for 1 year could lead to a significant improvement in DFS (p<0.0001), but there was no apparent improvement in OS (p=0·11). In our study, among the 259 patients only 33 (12.7%) received Trastuzumab therapy; Trastuzumab was administered concurrently with chemotherapy in 1 patient, and other 32 were treated sequentially after chemotherapy. In patients treated with Trastuzumab, when compared with those without Trastuzumab, 5-year OS increased from 77.6% to 93.8% (P=0.091), and 5-year DFS increased from 70.2% to 84.4% (P=0.09). Although no statistical difference was found between the two arms, it showed a trend

toward improvement in OS and DFS. And in the stratified analysis, neither were there any statistical differences in 5-year OS and DFS between 1-year (21cases) and 2-year (11 cases) Trastuzumab therapy. This might be attributed to the fact that there were too few cases enrolled and rather limited follow-up duration in these 2 subgroups.

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