ORIGINAL ARTICLE

Association between Pathological Complete Response and Outcome Following Neoadjuvant Chemotherapy in Locally Advanced Breast Cancer Patients

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Purpose: We aimed to determine the rate of pathological complete response (pCR), clinicopathological factors associated with pCR, and clinical outcomes following neoadjuvant chemotherapy in locally advanced breast cancer. **Methods:** Medical records of patients who had undergone neoadjuvant chemotherapy for breast cancer between January 2007 and September 2011 were retrospectively reviewed, and the pCR rates were calculated according to three sets of criteria: the National Surgical Adjuvant Breast and Bowel Project (NSABP), the MD Anderson Cancer Center (MDACC), and the German Breast Group (GBG). Tumors were classified as luminal A like, luminal B like, human epidermal growth factor receptor 2 (HER2), or triple-negative. pCR and clinical outcome, including overall survival (OS) and disease-free survival (DFS) rates were analyzed at the median follow-up of 54.2 months. **Results:** Of a total of 179 patients who had received neoadjuvant chemotherapy, 167 patients (93.3%) had locally advanced breast cancer and 12 patients (6.7%) had early-stage breast cancer. The majority of patients (152 patients, 89.4%) received anthracycline-based neoadjuvant chemotherapy. The ob-

jective clinical response rate was 61.5%, comprising clinical partial response in 5.5% and clinical complete response in 3.9% of patients. Twenty-one (11.7%), 20 (11.2%), and 17 patients (9.5%) achieved pCR according to NSABP, MDACC, and GBG definitions, respectively. pCR rates, as defined by NSABP, according to breast cancer subtype were 4.4%, 9.7%, 24.2%, and 19.2% in luminal A like, luminal B like, HER2, and triple-negative subtypes, respectively. Patients who achieved pCR had significantly better DFS (5-year DFS rates, 80% vs. 53%, $p = 0.030$) and OS (5-year OS rates, 86% vs. 54%, $p = 0.042$) than those who did not. **Conclusion:** The pCR rate following neoadjuvant chemotherapy for breast cancer in Thai women attending our institution was 11.7%; pCR was more frequently observed in HER2 and triple-negative breast tumor subtypes. Patients who achieved pCR had significantly improved survival.

Key Words: *Antineoplastic combined chemotherapy protocols, Breast neoplasms, Neoadjuvant therapy, Surgery, Treatment outcome*

INTRODUCTION

Breast cancer is the most common cancer and the leading cause of cancer-related mortality in women, both worldwide and in Thailand. Locally advanced breast cancer (LABC), the most advanced stage of nonmetastatic breast cancer, has a substantial risk of recurrence, metastasis, and death, with a 5-year overall survival (OS) rate of approximately 57% [1]. LABC includes patients with any tumor > 5 cm, or that involves the skin or chest wall, and also those with fixed axillary

lymph nodes, or ipsilateral supraclavicular, infraclavicular, or internal mammary nodal involvement [2]. LABC accounts for only 5% to 7% of all breast cancer in the United States [1], whereas it represents 24% to 27% of all newly diagnosed breast cancer cases in Thailand [3]. Despite multimodality treatment using systemic chemotherapy, surgery, and radiotherapy, the majority of patients develop metastases; therefore, LABC remains a clinical challenge.

Neoadjuvant chemotherapy is the standard treatment for locally advanced and inflammatory breast cancer, with the aim of achieving tumor resectability, as well as for patients with early breast cancer who are considering breast-conserving surgery (BCS). Neoadjuvant chemotherapy is advantageous because it shrinks tumors, thereby rendering inoperable tumors resectable; increases rates of BCS; enables early treatment of micrometastasis; and facilitates *in vivo* assessment of

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chemotherapy-sensitivity [4]. However, well-validated accurate pathological tumor staging cannot be performed after neoadjuvant therapy. As tumor progression occurs very rarely (1%–2%) after neoadjuvant therapy, operable tumors rarely become unresectable.

Patients treated with neoadjuvant chemotherapy achieve a clinical response in 50% to 80% of cases, with a clinical complete response (cCR) rate of 10% to 20% and a pathological complete response (pCR) rate to chemotherapy of 10%–30% [5-11]. As patients who achieve pCR have superior long-term outcomes, pCR is a potential surrogate marker of survival [12].

We performed a retrospective analysis to determine the rate of pCR, clinicopathological factors associated with pCR, and clinical outcomes in breast cancer patients treated with neoadjuvant chemotherapy.

METHODS

In this retrospective study, medical records of patients with nonmetastatic breast cancer, treated with neoadjuvant chemotherapy at Siriraj Hospital between January 2007 and September 2011, were reviewed. Patients' medical records were selected from the hospital database using ICD-10 coding. Only patients who had been treated with neoadjuvant chemotherapy and had undergone subsequent surgery at Siriraj Hospital, and had received postoperative chemotherapy, radiotherapy, and hormonal treatment, if indicated, were included in the present study. The study protocol was approved by Siriraj Institutional Review Board (protocol number: 222/2556[EC4]), Siriraj Hospital Faculty of Medicine, Mahidol University, Thailand.

Invasive breast cancer was diagnosed from core biopsies, and staging was performed according to the American Joint Committee on Cancer/Union for International Cancer Control (AJCC/UICC) tumor-node-metastasis (TNM) staging criteria (v.3 2010). Initial workups for distant metastases included chest radiography, liver ultrasonography, and bone scans.

Neoadjuvant and adjuvant chemotherapy regimens administered included: (1) AC (60 mg/m^2 doxorubicin and 600 mg/m^2 cyclophosphamide intravenously on day 1, every 3 weeks, for four cycles); (2) EC (90 mg/m^2 epirubicin and 600 mg/m^2 cyclophosphamide intravenously on day 1, every 3 weeks, for four cycles); (3) FAC (500 mg/m^2 fluorouracil, 50 mg/m^2 doxorubicin, and 500 mg/m^2 cyclophosphamide intravenously on day 1, every 3 weeks, for six cycles); (4) FEC (500 mg/m^2 fluorouracil, 90 mg/m^2 epirubicin, and 500 mg/m^2 cyclophosphamide intravenously on day 1, every 3 weeks, for six cycles); (5) CMF (100 mg/m^2/day cyclophosphamide orally on days 1–14, and 40 mg/m^2 methotrexate and 500 mg/m^2 fluorouracil intravenously on days 1 and 8, every 4 weeks, for six cycles); (6) GC

(1,000 mg/m^2 gemcitabine intravenously on days 1 and 8, and carboplatin AUC5 intravenously on day 1, every 3 weeks, for six cycles, as part of a clinical study [13]); and (7) D-FEC (75 mg/m^2 docetaxel intravenously on day 1, every 3 weeks, for three cycles, followed by 600 mg/m^2 fluorouracil, 90 mg/m^2 epirubicin, and 600 mg/m^2 cyclophosphamide intravenously on day 1, every 3 weeks, for three cycles). Trastuzumab (8 mg/kg loading dose, followed by 6 mg/kg every 3 weeks, for 1 year) was administered to patients with human epidermal growth factor receptor 2 (HER2) overexpressed.

Surgical procedures consisted of mastectomy or BCS. Adjuvant breast radiotherapy was administered to patients who had undergone BCS, as well as to patients with initial clinical stage cT3–T4 and cN2–3 disease. Adjuvant endocrine therapy was administered to all patients with hormone receptor-positive tumors for 5 years.

Assessment of clinical response

The clinical response was assessed following administration of the final neoadjuvant chemotherapy cycle. The following definitions were used [6]: cCR was defined as the absence of clinically evident tumor on palpation; clinically partial response (cPR) was defined as a reduction of 50% or more in the two maximum perpendicular diameters of the tumor; clinically progressive disease (cPD) was defined as an increase of >25% in the two maximum perpendicular diameters of the tumor; and clinically stable disease (cSD) was defined as a clinical breast response that does not meet the definitions of cCR, cPR, or cPD.

Pathological assessment

Expression of estrogen receptor (ER), progesterone receptor (PR), and HER2 was determined on pretreatment biopsies (preferentially) or on surgical specimens if immunohistochemistry (IHC) had not previously been performed. Hormonal receptor (HR) status was considered positive if ≥1% of tumor cells stained for ER and/or PR. HER2 status was considered positive if an IHC score of 3+ was recorded, or if there was positive gene amplification using *in situ* hybridization testing. As Ki-67 assessment had not been routinely performed, it was not possible to define breast cancer intrinsic subtypes according to the St. Gallen International Expert Consensus on the Primary Therapy of Early Breast Cancer [14]. Accordingly, histological grade was used to rank cell proliferation. The following definitions of tumor types were used [15]: (1) luminal A like-tumors, defined as ER-positive and/or PR-positive, HER2-negative, grade 1 or 2; (2) luminal B like-tumors, defined as ER-positive and/or PR-positive, HER2-negative, grade 3; or ER-positive and/or PR-positive, HER2-positive, all grades; (3)

HER2-like tumors, defined as ER- and PR-negative, HER2-positive, all grades; and (4) triple-negative tumors: ER-, PR-, and HER2-negative, all grades.

pCR was evaluated according to the criteria [15] of the National Surgical Adjuvant Breast and Bowel Project (NSABP), the MD Anderson Cancer Center (MDACC), and the German Breast Group (GBG), as no invasive cancer in the breast (ypT0/is ypN0/+), no invasive cancer in the breast and lymph nodes (ypT0/is ypN0), and no invasive or *in situ* cancer in the breast and lymph nodes (ypT0 ypN0), respectively.

Statistical assessment

The primary endpoint of this study was to determine the rate of pCR. The secondary endpoints were to determine the clinical factors associated with pCR, the clinical response rate, disease-free survival (DFS; defined as the interval between the date of diagnosis and the date of disease recurrence or death), and OS (defined as the interval between the date of diagnosis and the date of death from any cause). The required sample size was calculated based on an estimated proportion of one group method, using 13% for pCR in accordance with previous NSABP studies [5,7], with a 95% confidence interval (CI) and 5% error; this calculation resulted in a required sample size of 174 patients. On univariate analysis, the relationships between clinical factors and pCR were assessed using Pearson chi-square or Fisher exact test, as appropriate, and binary logistic regression was used to calculate the odds ratio.

Survival was analyzed using the Kaplan-Meier method, and comparisons made using the log-rank or Breslow test. The Cox proportional hazard model for survival was used for univariate and multivariate analyses. Median follow-up time was calculated using the reverse Kaplan-Meier method. A two-sided level of significance of 0.05 was applied to all statistical tests. SPSS Statistical software version 19.0 (IBM Corp., Armonk, USA) was used for all statistical analyses.

RESULTS

Patients and tumor characteristics

The total number of patients treated with chemotherapy followed by surgery between January 1, 2007 and September 31, 2011 was 237. Data of 179 patients met the inclusion criteria and were used in this study (Figure 1). The median patient age was 48 years (range, 24–75 years), and 59.8% of patients were premenopausal. LABC was diagnosed in 93.3% of cases. The mean tumor size on palpation was 7.9 cm. The majority of patients (69.3%) had node-positive disease. Histological examination revealed that 94.4% of tumors were invasive ductal carcinoma. Tumors were HR-positive in 63.1% of cases (ER-posi-
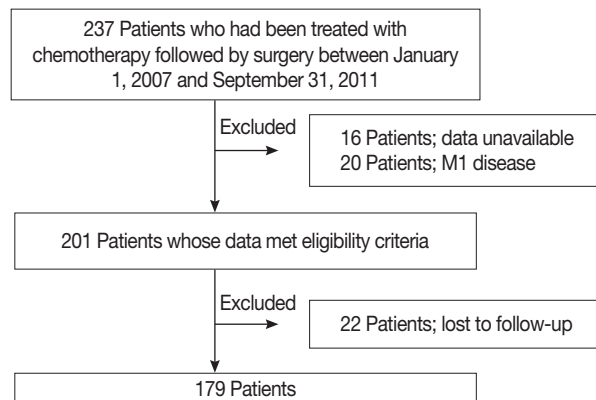

Figure 1. Consort diagram. There were 237 patients who treated with chemotherapy followed by surgery between January 1, 2007 and September 31, 2011. Data of 179 patients met the inclusion criteria and were used in this study.

tive, 58.1%; PR-positive, 50.8%). Tumors were HER2-positive in 39.7%, HER2-negative in 50.8%, and equivocal in 9.5%. IHC results were reported from preoperative specimens in 71.5%, and postoperative specimens in 28.5%. Of the 179 tumors, 166 (92.7%) could be classified into one of the four intrinsic subtypes: luminal A like (25.1%), luminal B like (34.6%), HER2 (18.4%), and triple-negative (14.5%).

In all, 152 (89.4%) received anthracycline-based chemotherapy, AC, EC, FAC, or FEC regimens. Of the 71 patients with HER2-positive tumors, 19 (26.8%) received neoadjuvant trastuzumab. At least four cycles of neoadjuvant chemotherapy were received by 66.5% patients. Total mastectomy was performed on 162 (90.5%) of the patients, and the remaining 17 (9.5%) underwent BCS. Patients and tumor characteristics are shown in Table 1.

pCR and clinical objective response

cCR and cPR were observed in seven (3.9%) and 103 (57.5%) patients, resulting in a total clinical response rate of 61.5% (Table 2). Clinical progression was seen in two patients (1.1%). Among the 69 nonresponders (cSD and cPD), seven tumors remained inoperable, and these patients received additional neoadjuvant therapy. Five of these patients were treated with a second neoadjuvant chemotherapy regimen (mostly taxane-based regimens), and one patient received preoperative radiotherapy. The remaining patient received both a second neoadjuvant chemotherapy regimen and preoperative breast radiation. All seven patients eventually underwent total mastectomy. The absence of invasive cancer in the breast (ypT0) was found in 19 patients (10.6%), and the absence of cancer in the lymph nodes (ypN0) was found in 57 (31.8%). pCR occurred in 21 (11.7%), 20 (11.2%), and 17 patients (9.5%), according to the NSABP, MDACC, and GBG criteria, respectively (Table 2).

Table 1. Patients' characteristics

Characteristic	No. (%)
Age at diagnosis (yr)*	48 (24–75)
Menopausal status	
Premenopause	107 (59.8)
Postmenopause	60 (33.5)
Unknown	12 (6.7)
Reason for neoadjuvant	
Locally advanced	167 (93.3)
Goal for BCS	12 (6.7)
Histology type	
Ductal invasive	169 (94.4)
Lobular invasive	10 (5.6)
Clinical T stage	
cTx-1	5 (2.8)
cT2	14 (7.8)
cT3	68 (38.0)
cT4	92 (51.4)
Tumor size (cm)*	7.9 (1.6–20)
Clinical N stage	
N (–)	55 (30.7)
N (+)	124 (69.3)
Tumor grade	
1	9 (5.0)
2	76 (42.5)
3	82 (45.8)
Unknown	12 (6.7)
ER status	
Positive	104 (58.1)
Negative	75 (41.9)
PR status	
Positive	91 (50.8)
Negative	88 (49.2)
HER2 status	
Positive	71 (39.7)
Negative	91 (50.8)
Equivocal	17 (9.5)
Intrinsic breast subtype	
Luminal A like	45 (25.1)
Luminal B like	62 (34.6)
HER2	33 (18.4)
Triple-negative	26 (14.5)
Unknown	13 (7.3)
Neoadjuvant chemotherapy	
AC or EC	71 (41.8)
FAC or FEC	81 (47.6)
CMF	5 (2.9)
GC	3 (1.8)
Chemotherapy+Trastuzumab	19 (11.2)
No. of cycles	
≤3	60 (33.5)
>3	119 (66.5)
Surgery type	
Total mastectomy	162 (90.5)
BCS	17 (9.5)

BCS=breast-conserving surgery; ER=estrogen receptor; PR=progesterone receptor; HER2=human epidermal growth factor receptor 2; AC=doxorubicin+cyclophosphamide; EC=epirubicin+cyclophosphamide; FAC=fluorouracil+doxorubicin+cyclophosphamide; FEC=fluorouracil+epirubicin+cyclophosphamide; CMF=cyclophosphamide+methotrexate+fluorouracil; GC=gemcitabine+carboplatin.
*Median (range).

Table 2. Clinical and pathological responses to neoadjuvant chemotherapy

Clinical and pathological response	No. (%)
Clinical response	
cCR	7 (3.9)
cPR (>50%)	103 (57.5)
cSD	67 (37.4)
cPD (>25%)	2 (1.1)
Objective response rate	110 (61.5)
Pathological tumor stage	
ypT0	19 (10.6)
ypTis	7 (3.9)
ypT1–4	153 (85.4)
Pathological N stage	
ypN0	57 (31.8)
ypN1	55 (30.7)
ypN2–3	67 (37.5)
Pathological complete response	
NSABP criteria	21 (11.7)
MDACC criteria	20 (11.2)
German criteria	17 (9.5)

cCR=clinically complete response; cPR=clinically partial response; cSD=clinically stable disease; cPD=clinically progressive disease; NSABP=National Surgical Adjuvant Breast and Bowel Project; MDACC=MD Anderson Cancer Center.

Association between baseline clinicopathological factors and pCR using the NSABP criteria

According to univariate analysis, pCR was significantly associated with HR status and intrinsic breast cancer subtype (Table 3). The pCR rate was lower in HR-positive tumors (defined as ER- and/or PR-positive) than in HR-negative tumors (7.1% vs. 19.7%; 95% CI, 1.26–8.25; $p=0.015$). However, only ER status had a statistically significant association with pCR rate (pCR was 21.3% for ER-negative tumors and 4.8% for ER-positive tumors; 95% CI, 1.87–15.42; $p=0.002$). Analysis of the four intrinsic breast cancer subtypes found that pCR rates were 4.4%, 9.7%, 24.2%, and 19.2% for luminal A like, luminal B like, HER2, and triple-negative tumors, respectively. The pCR rate was significantly higher in the HER2 subtype than that in the luminal A like subtype (95% CI, 1.35–34.97; $p=0.020$). However, on multivariate analysis, only ER-negative tumors were significantly associated with pCR (95% CI, 1.32–48.2; $p=0.024$) (Table 4).

Treatment following neoadjuvant chemotherapy and surgery

For 62 patients (35%), postoperative chemotherapy regimens were changed to taxane-based (60 patients) or CMF regimens (two patients), whereas 117 patients (65%) continued with the same chemotherapy regimen that they had received preoperatively (predominantly anthracycline-based). Adjuvant endocrine therapy and trastuzumab were adminis-

Table 3. Association between clinicopathological factors and pathological complete response rate (NSABP criteria)

Factor	No.	pCR No. (%)	Univariate analysis OR (95% CI)	p-value
Age (yr)				
>50	70	9 (12.9)	1.19 (0.47–3.00)	0.708
≤50	109	12 (11.0)	1.00	
Menopausal status				
Postmenopause	60	9 (15.0)	1.71 (0.65–4.48)	0.270
Premenopause	107	10 (9.3)	1.00	
Reason for neoadjuvant				
Goal BCS	12	2 (16.7)	1.97 (0.39–9.99)	0.411
Locally advanced	167	19 (11.4)	1.00	
Clinical tumor stage				
cT0–3	87	12 (13.8)	1.48 (0.59–3.70)	0.407
cT4	92	9 (9.8)	1.00	
Clinical nodal stage				
Negative	55	7 (12.7)	1.15 (0.43–3.02)	0.783
Positive	124	14 (11.3)	1.00	
Tumor grade				
3	82	13 (15.9)	1.81 (0.71–4.64)	0.209
1 or 2	85	8 (9.4)	1.00	
ER status				
Negative	75	16 (21.3)	5.37 (1.87–15.42)	0.002
Positive	104	5 (4.8)	1.00	
PR status				
Negative	88	14 (15.9)	2.27 (0.87–5.93)	0.094
Positive	91	7 (7.7)	1.00	
Hormornal receptor status				
Negative	66	13 (19.7)	3.22 (1.26–8.25)	0.015
Positive	113	8 (7.1)	1.00	
HER2 status				
Positive	71	11 (15.5)	1.49 (0.59–3.72)	0.399
Negative	91	10 (11.0)	1.00	
Equivocal	17	0		
Intrinsic breast subtype				
Luminal A-like	45	2 (4.4)	1.00	
Luminal B-like	62	6 (9.7)	2.30 (0.44–11.98)	0.321
HER2	33	8 (24.2)	6.88 (1.35–34.97)	0.020
Triple-negative	26	5 (19.2)	5.12 (0.92–28.61)	0.063
Neoadjuvant chemotherapy				
AC or EC	71	8 (11.3)	1.00	
FAC or FEC	81	6 (7.4)	0.63 (0.21–1.91)	0.415
Anti-HER2+CMT	18	7 (38.9)	5.01 (1.51–16.63)	0.008
Others	8	0		
No. of neoadjuvant cycles				
≤3	60	4 (6.7)	1.00	
>3	119	17 (14.3)	2.33 (0.75–7.27)	0.144

NSABP=National Surgical Adjuvant Breast and Bowel Project; pCR=pathological complete response; OR=odds ratio; CI=confidence interval; BCS=breast-conserving surgery; ER=estrogen receptor; PR=progesterone receptor; HER2=human epidermal growth factor receptor 2; AC=doxorubicin+cyclophosphamide; EC=epirubicin+cyclophosphamide; FAC=fluorouracil+doxorubicin+cyclophosphamide; FEC=fluorouracil+epirubicin+cyclophosphamide; CMT=chemotherapy.

Table 4. Multivariate analysis of factors possibly associated with pathological complete response

Factor	Multivariate analysis OR (95% CI)	p-value
Tumor grade		
3	1.14 (0.37–3.49)	0.817
1 or 2		
ER status		
Negative	7.97 (1.32–48.20)	0.024
Positive		
PR status		
Negative	0.89 (0.09–8.62)	0.919
Positive		
HR status		
Negative	0.71 (0.02–21.03)	0.841
Positive		
Intrinsic breast subtype		
Luminal A-like		
Luminal B-like	1.40 (0.21–9.37)	0.732
HER2	0.99 (0.04–27.67)	0.997
Triple-negative	0.71 (0.02–21.03)	0.841
No. of neoadjuvant cycles		
≤3		
>3	2.23 (0.67–7.36)	0.189

CI=confidence interval; ER=estrogen receptor; PR=progesterone receptor; HR=hormonal receptor; HER2=human epidermal growth factor receptor 2.

tered to 113 (63%) and 30 patients (17%), respectively. The majority of patients (94%) also received postoperative radiotherapy (Supplementary Table 1).

Survival

The median follow-up duration of the 179 patients was 54.2 months. At the cutoff date for follow-up (September 20, 2013), 59 patients (33%) had disease recurrence, and 55 patients had died (30.7%). Of the 59 patients with recurrent disease, 47 (80%) had distant metastases, seven (11%) had local relapse, and five (9%) had contralateral breast cancer occurrence. The 5-year DFS and OS rates of all patients in this study were 56% and 57%, respectively.

Univariate analysis was performed using the Cox regression method to evaluate whether relevant clinicopathological variables, and known prognostic factors had significant associations with DFS and OS (Table 5). Factors that were significantly associated with improved DFS and OS were low-to-moderate tumor grade, ER positivity of >50%, PR positivity, luminal A like subtype, ypT0/is, ypN0-1, and absence of angiolymphatic invasion. pCR was predictive of longer DFS (95% CI, 0.09–0.95; p=0.041), and trended towards longer OS (95% CI, 0.06–1.05; p=0.059).

Compared with luminal A like subtype, luminal B like tumors were significantly associated with a shorter DFS (unad-

Table 5. Univariate analysis (Cox regression) of effects of assessed factors on disease-free survival and overall survival

Factor	No.	Event No. (%)	DFS HR (95% CI)	DFS p-value	Death No. (%)	OS HR (95% CI)	OS p-value
Age (yr)							
≤50	109	45 (39.0)	1.64 (0.97–2.75)	0.063	36 (33)	1.40 (0.80–2.44)	0.239
>50	70	21 (26.9)	1.00		19 (27.1)	1.00	
Tumor grade							
1 or 2	85	24 (28.2)	1.00		19 (22.4)	1.00	
3	82	40 (48.8)	1.78 (1.07–2.95)	0.026	34 (41.5)	1.87 (1.06–3.27)	0.030
ER							
Negative	75	32 (42.7)	1.56 (0.96–2.52)	0.073	28 (37.3)	1.64 (0.97–2.79)	0.067
Positive	104	34 (32.7)	1.00		27 (26.0)	1.00	
% ER							
Negative	75	32 (42.7)	1.90 (1.11–3.25)	0.019	28 (37.3)	2.11 (1.15–3.86)	0.015
Positive <50%	20	11 (55.0)	2.24 (1.09–4.60)	0.028	10 (50.0)	2.47 (1.13–5.42)	0.023
Positive >50%	84	23 (27.4)	1.00		17 (20.2)	1.00	
PR							
Negative	88	39 (44.3)	1.69 (1.03–2.76)	0.036	34 (38.6)	1.74 (1.01–2.99)	0.047
Positive	91	27 (29.7)	1.00		21 (23.1)	1.00	
HER2							
Negative	91	34 (37.4)	1.00		28 (30.8)	1.00	
Positive	71	28 (39.4)	0.50 (0.18–1.41)	0.190	23 (32.4)	0.99 (0.57–1.72)	0.971
Equivocal	17	4 (23.5)	1.11 (0.67–1.83)	0.688	4 (23.5)	0.53 (0.19–1.52)	0.240
Intrinsic breast subtype							
Luminal A-like	45	8 (17.8)	1.00		7 (15.6)	1.00	
Luminal B-like	62	27 (43.5)	2.74 (1.24–6.03)	0.012	20 (32.3)	2.04 (0.86–4.84)	0.104
HER2	33	14 (42.4)	3.05 (1.28–7.28)	0.012	13 (39.4)	2.86 (1.14–7.16)	0.025
Triple-negative	26	13 (50.0)	3.42 (1.42–8.28)	0.006	11 (42.3)	2.96 (1.14–7.67)	0.026
ypT staging							
ypT0/is	26	4 (15.4)	1.00		2 (7.7)	1.00	
ypT1–4	153	62 (40.5)	3.19 (1.16–8.76)	0.025	53 (34.6)	5.18 (1.26–21.26)	0.022
ypN staging							
ypN0	57	17 (29.8)	1.00		14 (24.6)	1.00	
ypN1	55	12 (21.8)	0.74 (0.35–1.54)	0.418	9 (16.4)	0.73 (0.32–1.69)	0.461
ypN2	35	19 (54.3)	2.34 (1.21–4.51)	0.011	15 (42.9)	2.35 (1.13–4.88)	0.022
ypN3	32	18 (56.3)	2.77 (1.42–5.40)	0.003	17 (53.1)	3.92 (1.92–8.03)	0.000
pCR by NSABP							
Yes	21	3 (14.3)	0.30 (0.09–0.95)	0.041	2 (9.5)	0.26 (0.06–1.05)	0.059
No	158	63 (39.9)	1.00		53 (33.5)	1.00	
ALI							
No	80	29 (36.3)	1.00		26 (32.5)	1.00	
Yes	68	34 (50.0)	1.84 (1.12–3.03)	0.017	27 (39.7)	1.81 (1.05–3.13)	0.033

DFS = disease-free survival; OS = overall survival; HR = hazard ratio; CI = confidence interval; ER = estrogen receptor; PR = progesterone receptor; HER2 = human epidermal growth factor receptor 2; yp = posttreatment pathologic findings; pCR = pathological complete response; NSABP = National Surgical Adjuvant Breast and Bowel Project; ALI = angiolymphatic invasion.

justed hazard ratio [HR], 2.74; *p* = 0.012) and nonsignificantly with a shorter OS (unadjusted HR, 2.04; *p* = 0.104), whereas HER2 and triple-negative tumors were significantly associated with shorter DFS and OS (HER2 tumors: unadjusted HR for DFS, 3.05, *p* = 0.012; unadjusted HR for OS, 2.86, *p* = 0.025; triple-negative: unadjusted HR for DFS, 3.42, *p* = 0.006; unadjusted HR for OS, 2.96, *p* = 0.026) (Figure 2, which shows Kaplan-Meier survival curves).

Kaplan-Meier survival curves according to pCR are shown in Figure 3. Patients who achieved pCR showed significant positive associations with DFS and OS compared to those without a pCR (5-year DFS: 80% vs. 53%, log-rank test, *p* = 0.030; 5-year OS: 86% vs. 54%, log-rank test, *p* = 0.042). Subgroup analysis according to ER status demonstrated that pCR was significantly associated with longer DFS (*p* = 0.007) and OS (*p* = 0.004) in patients with ER-negative tumors. In contrast, there was no difference in survival outcome between patients with ER-positive tumors with or without pCR (Supple-
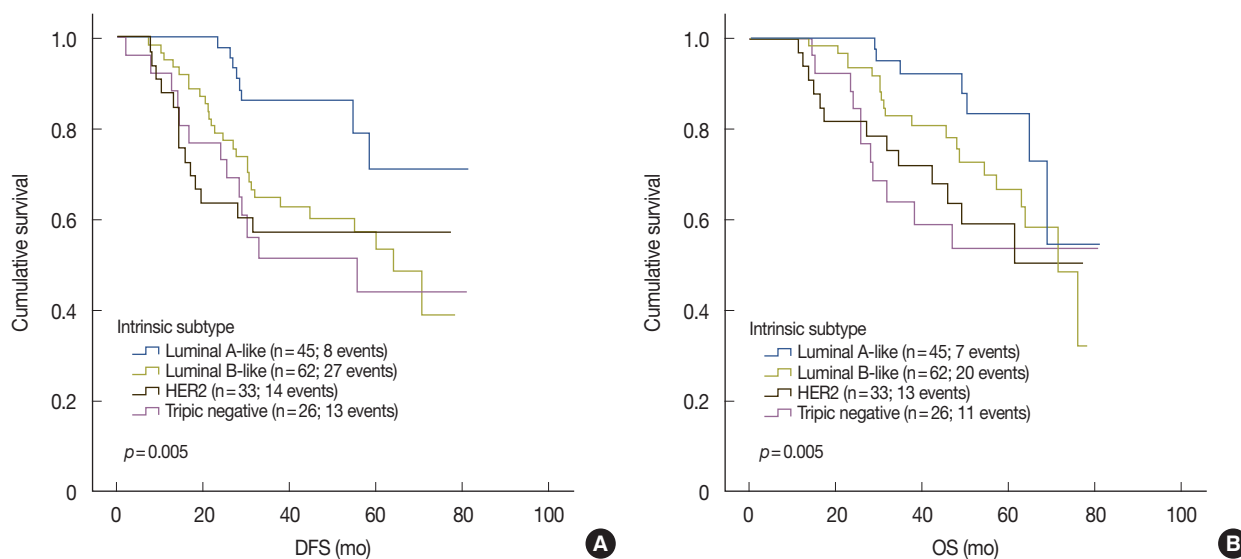
Figure 2. Disease-free survival (DFS) and overall survival (OS) according to intrinsic breast cancer subtypes. Compared with luminal A like tumor, luminal B like tumors, human epidermal growth factor receptor 2 (HER2) and triple-negative tumors were associated with a shorter DFS (A) and OS (B).
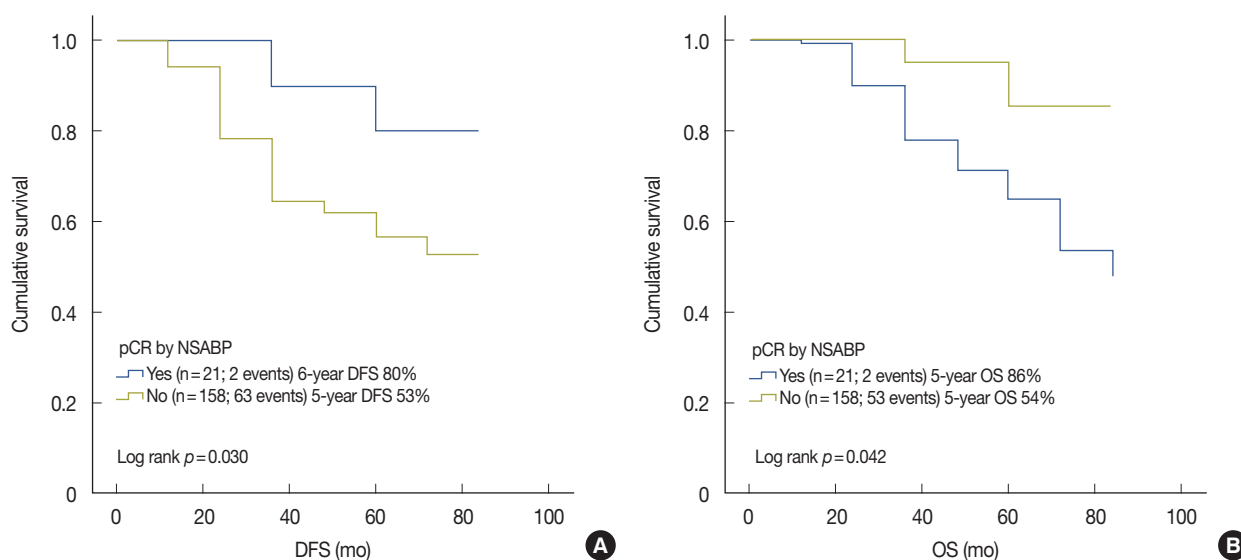

Figure 3. Disease-free survival (DFS) and overall survival (OS) according to pathological complete response (pCR). Patients who achieved pCR showed significant positive associations with DFS (A) and OS (B) compared to those without a pCR (5-year DFS: 80% vs. 53%, log-rank test, $p=0.030$; 5-year OS: 86% vs. 54%, log-rank test, $p=0.042$).
NSABP=National Surgical Adjuvant Breast and Bowel Project.

mentary Figure 1).

In this study, ER status (ER-positive vs. ER-negative) was not significantly associated with survival outcome. However, ER-negative tumors showed a general trend towards shorter DFS and OS than ER-positive tumors, as can be seen from the Kaplan-Meier survival curves. The potential influence of the strength of ER-positivity was therefore explored, and patients with ER positivity < 50% had worse prognoses (DFS, $p=0.028$; OS, $p=0.023$) as the same as ER-negative tumors (DFS, $p=0.019$; OS, $p=0.015$). Conversely, tumors with ER positivity

> 50% had significantly better prognoses (Supplementary Figure 2).

According to multivariate Cox proportional hazard regression analysis, the independent risk factors that were significantly associated with prolonged DFS and OS were pCR and intrinsic breast cancer subtypes; HER2 and triple-negative subtypes were independent risk factors for poorer outcomes (Table 6).

Table 6. Multivariate Cox hazard regression analysis of disease-free survival and overall survival

Factor	No.	Event No. (%)	DFS		Death No. (%)	OS	
			HR (95% CI)	p-value		HR (95% CI)	p-value
pCR by NSABP							
Yes	21	3 (14.3)	0.28 (0.08–0.95)	0.041	2 (9.5)	0.22 (0.05–0.97)	0.045
No	158	63 (39.9)	1.00		53 (33.5)	1.00	
Intrinsic breast subtype							
Luminal A-like	45	8 (17.8)	1.00		7 (15.6)	1.00	
Luminal B-like	62	27 (43.5)	2.15 (0.97–4.78)	0.059	20 (32.3)	1.69 (0.71–4.03)	0.235
HER2	33	14 (42.2)	3.61 (1.49–8.76)	0.004	13 (39.4)	3.40 (1.34–8.66)	0.010
Triple-negative	26	13 (50.0)	3.50 (1.39–8.80)	0.008	11 (42.3)	3.08 (1.12–8.46)	0.029
ALI							
No	80	29 (36.3)	1.00		26 (32.5)	1.00	
Yes	68	34 (50.0)	1.66 (0.96–2.89)	0.070	27 (39.7)	1.68 (0.91–3.12)	0.098
Tumor grade							
1 or 2	85	24 (28.2)	1.00		19 (22.4)	1.00	
3	82	40 (48.4)	0.87 (0.46–1.63)	0.662	34 (41.5)	1.25 (0.61–2.56)	0.542

DFS=disease-free survival; OS=overall survival; HR=hazard ratio; CI=confidence interval; pCR=pathological complete response; NSABP=National Surgical Adjuvant Breast and Bowel Project; HER2=human epidermal growth factor receptor 2; ALI=angiolymphatic invasion.

DISCUSSION

Neoadjuvant chemotherapy is the standard treatment for locally advanced and inflammatory breast cancer, and is also offered to patients with early breast cancer who are considering BCS. In developed countries, BCS is used in most cases of breast cancer. However, in Thailand, the majority of patients present with locally advanced disease, making the main aim of initial treatment to downstage the disease and render inoperable tumors resectable. In the present study, the pCR rate following neoadjuvant chemotherapy was 11.7%. pCR was more frequently observed in HER2 and triple-negative breast tumor subtypes. Patients who achieved pCR had significantly improved survival.

At present, the most widely accepted criteria to measure response to chemotherapy are the Response Evaluation Criteria for Solid Tumors (RECIST). However, in this study, we used World Health Organization (WHO) criteria to enable comparison of our findings with those of previously published studies of neoadjuvant chemotherapy, most of which have used WHO criteria [5-9]. Anthracycline-based regimens are highly effective in breast cancer and have showed to result in pCR rates of 10% to 15% [5,8]. The addition of taxane to anthracycline-based regimens was shown to increase the pCR rate to 25% to 30%, but did not have an impact on DFS or OS [7,9,16]. In the two large randomized NSABP studies, NSABP B-18 [5], and B-27 [7], four cycles of an AC regimen achieved a pCR rate of 13%; our pCR rate of 11% was slightly lower. This may be attributable to the greater proportion of patients with locally advanced stage disease in our study; most patients

in the NSABP studies had earlier, operable breast cancer (Supplementary Table 2) [5-11,17-20]. In a retrospective MDACC study [17], 372 patients with LABC were treated with four cycles of neoadjuvant doxorubicin-containing regimens, mainly FAC, and this resulted in a pCR rate of 12%, which is also consistent with our current study. In contrast, patients with operable breast cancer in the European Organisation for Research and Treatment of Cancer 10902 trial [8] received four cycles of neoadjuvant FEC regimens and achieved only a 4% pCR rate. Another clinical series [18] of 110 LABC patients received between three and eight cycles (mean, 4) of neoadjuvant anthracycline and/or taxane-containing chemotherapy, and reported a pCR rate of 5.5%. The lower pCR rates seen in these trials may be explained by the higher proportion of patients with ER-positive tumors, which are considered to be less responsive to chemotherapy.

Previous studies have addressed clinical and biological factors associated with pCR following neoadjuvant chemotherapy and demonstrated that breast cancer patients with high tumor grade or ER-negative disease [17,21-23], and HER2 breast cancer treated with trastuzumab combined with chemotherapy [24-26], were more likely to achieve pCR. In the present study, we calculated the pCR rate using NSABP criteria so that we could compare this rate with other clinical and biological factors. We also reported the pCR rates using all three established sets of criteria, to allow comparison of these rates reported in other studies that may have utilized different criteria. ER-negative tumor status, but not tumor grade, was found to be a predictor of pCR. HER2-positive tumor status was not associated with a higher pCR rate, and this may be because the majority

of these patients (75%) did not received anti-HER2 therapy. When only patients treated with neoadjuvant chemotherapy combined with anti-HER2 therapy were analyzed, a higher pCR rate was observed.

Patients achieving pCR in this study had 5-year DFS and OS rates of 80% and 86%, respectively, compared to those of 53% and 54% for those with non-pCR. The significantly improved survival in patients with pCR is in concordance with most neo-adjuvant chemotherapy trials [5,8,10,16,17]. In our study, some of the patients were also treated with adjuvant chemotherapy, which might have an effect on survival outcome. Therefore, we further explored the effect of adjuvant chemotherapy on survival, but did not find any significant effect.

According to univariate analysis, clinicopathological factors associated with prolonged survival outcome in this study were low-to-moderate tumor grade, absence of angiolymphatic invasion, positive PR status, ER-positivity of > 50%, and luminal-type cancer. Patients with luminal A like tumors had better prognoses than patients with luminal B like, HER2, or triple-negative tumors, despite having the lowest pCR rate (4%). Among the four intrinsic breast subtypes, the pCR rate was higher in the triple-negative (19%) and HER2 (24%) groups than in the luminal subtypes.

Although pCR was significantly associated with prolonged DFS, there was only borderline significance of association with OS. This could be explained by the finding that pCR mainly predicted survival outcome in ER-negative breast cancer, but not in ER-positive tumor. Therefore, pCR may be a good predictor of survival for nonluminal (ER-negative) disease following neoadjuvant chemotherapy, rather than for those with luminal breast cancer. Consistent with the findings of the present study, a recent meta-analysis of 6,377 breast cancer patients from seven randomized trials, demonstrated that pCR was associated with improved DFS in luminal B/HER2-negative, nonluminal HER2-positive, and triple-negative disease, but not in luminal A disease [15].

The limitations of this study are its retrospective nature and inclusion of patients from a single center. However, few randomized studies have focused on LABC. This may be attributable to its low incidence (5%–6%) in developed countries compared to developing countries, where LABC accounts for 30% of breast cancer.

The present retrospective study has demonstrated that treatment of Thai LABC patients with anthracycline-based neoadjuvant chemotherapy yields pCR rates comparable to those reported by randomized trials of patients with operable breast cancers in developed countries. pCR may be used as an positive prognostic indicator following neoadjuvant chemotherapy.

CONFLICT OF INTEREST

The authors declare that they have no competing interests.

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