Original Article

Analysis of Ki-67 Expression With Neoadjuvant Anastrozole or Tamoxifen in Patients Receiving Goserelin for Premenopausal Breast Cancer

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BACKGROUND: The increasing costs associated with large scale adjuvant trials mean that the prognostic value of biologic markers is increasingly important. The expression of nuclear antigen Ki 67, a marker of cell proliferation, has been correlated with treatment effi cacy and is being investigated for its value as a predictive marker of therapeutic response. In the current study, the authors explored correlations between Ki 67 expression and tumor response, estrogen receptor (ER) status, progesterone receptor (PgR) status, and histopathologic response from the STAGE study (Study of Tamoxifen or Arimidex, combined with Goserelin acetate to compare Effi cacy and safety). **METHODS:** In a phase 3, double blind, randomized trial (National Clinical Trials identifier NCT00605267), premeno pausal women with ER positive, early stage breast cancer received either anastrozole plus goserelin or tamoxifen plus goserelin for 24 weeks before surgery. The Ki 67 index, hormone receptor (ER and PgR) status, and histopathologic responses were determined from histopathologic samples that were obtained from core needle biopsies at baseline and at surgery. Tumor response was deter mined by using magnetic resonance imaging or computed tomography. **RESULTS:** In total, 197 patients were randomized to receive either anastrozole plus goserelin (n = 98) or tamoxifen plus goserelin (n = 99). The best overall tumor response was better for the anastrozole group compared with the tamoxifen group both among patients who had a baseline Ki 67 index ≥20% and among those who had a baseline Ki 67 index <20%. There was no apparent correlation between baseline ER status and the Ki 67 index in either group. Positive PgR status was reduced from baseline to week 24 in the anastrozole group. **CONCLUSIONS:** In premenopausal women with ER positive breast cancer, anastrozole produced a greater best overall tumor response compared with tamoxifen regard less of the baseline Ki 67 index. ***Cancer* 2013;119:704-13.** © *2012 American Cancer Society.*

KEYWORDS: anastrozole, aromatase inhibitor, biomarker, neoadjuvant, Ki 67, premenopausal breast cancer.

INTRODUCTION

In addition to ablative surgery, radiotherapy, and cytotoxic chemotherapy, an additional standard treatment option for premenopausal women with estrogen receptor (ER)-positive breast cancer is the ER antagonist tamoxifen, either alone or in combination with ovarian function suppression.[1] Temporary and potentially reversible ovarian suppression can be achieved by treatment with a luteinizing hormone-releasing hormone analog, such as goserelin. Goserelin in combination with tamoxifen has demonstrated improved progression-free survival and disease-free survival compared with goserelin alone in premenopausal women with hormone receptor-positive (ER-positive and/or progesterone receptor [PgR]-positive) breast cancer in the advanced[2] and adjuvant[3] settings.

Nonsteroidal aromatase inhibitors (AIs), including anastrozole and letrozole, and the irreversible steroidal aromatase inactivator exemestane have demonstrated improved efficacy compared with tamoxifen in the advanced[4-7] and adjuvant[8-

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[12] treatment settings. Therefore, AIs in combination with ovarian suppression have been evaluated for the treatment of premenopausal women with ER-positive breast cancer.[13,14]

Neoadjuvant treatment for breast cancer provides an opportunity for downstaging of large tumors to allow patients to undergo breast-conserving surgery rather than mastectomy. Chemotherapy can offer an effective neoadjuvant treatment; however, increasing evidence suggests that ER-positive tumors are less sensitive to chemotherapy.[15] It has been demonstrated that neoadjuvant endocrine therapy has efficacy in the treatment of ER-positive disease among postmenopausal women, resulting in similar objective response rates and rates of breast-conserving surgery for AIs compared with more cytotoxic chemotherapy.[16] Therefore, the role of neoadjuvant endocrine therapy in premenopausal women is also of interest.

With the increasing costs associated with large-scale adjuvant trials, both the prognostic value of biologic markers and the long-term predictive value of short-term trials are increasingly important. The expression of nuclear antigen Ki-67, a marker of cell proliferation, reportedly has been correlated with treatment efficacy and is being investigated for its value as a predictive marker of therapeutic response.[17] In a cross-trial comparison, an increased reduction in Ki-67 expression after neoadjuvant treatment with anastrozole compared with tamoxifen was observed consistently; and increased progression-free survival has been reported for anastrozole versus tamoxifen in the adjuvant Arimidex, Tamoxifen, Alone or in Combination (ATAC) trial.[8,18,19]

The STAGE study (Study of Tamoxifen or Arimidex Combined With Goserelin Acetate to Compare Efficacy and Safety) was the first randomized trial to compare anastrozole plus goserelin versus tamoxifen plus goserelin in the neoadjuvant setting (24 weeks of therapy) in premenopausal women with ER-positive and human epidermal growth factor receptor 2 (HER2)-negative, operable breast cancer. The patients who received anastrozole plus goserelin in that trial had a superior best overall tumor response compared with the patients who received tamoxifen plus goserelin, as measured on magnetic resonance imaging (MRI) or computed tomography (CT) studies (anastrozole plus goserelin, 64.3%; tamoxifen plus goserelin, 37.4%; estimated difference, 26.9%; 95% confidence interval [CI], 13.5-40.4; $P < .001$). The treatment effect was consistently in favor of anastrozole, regardless of the measurement methods (caliper and ultrasound). The histopathologic response rate also was better in the anastrozole group (anastrozole plus goserelin, 41.8%; tamoxifen plus goserelin, 27.3%; estimated difference, 14.6%; 95% CI, 1.4-27.7; $P = .032$). Both treatment regimens were well tolerated, consistent with the known safety profiles of anastrozole, tamoxifen, and goserelin.[20] The geometric mean Ki-67 index at baseline was 21.9% in the anastrozole group and 21.6% in the tamoxifen group. At week 24, the Ki-67 index was reduced in both treatment groups (to 2.9% in the anastrozole group and to 8% in the tamoxifen group). The reduction from baseline to week 24 was significantly greater with anastrozole than with tamoxifen. The estimated ratio of reduction between groups was 0.35 (95% CI, 0.24-0.51; $P < .001$).[20] Here, we report an exploratory analysis of the STAGE study that investigated potential correlations between the Ki-67 index and the best overall tumor response, ER status, PgR status, or histopathologic response.

MATERIALS AND METHODS

Study Design and Patients

In this phase 3, double-blind, randomized, parallel-group, multicenter trial, the participating patients were premenopausal women ≥20 years with ER-positive and HER2-negative breast cancer who had operable and measurable lesions (tumors measuring 2-5 cm, negative lymph node status [N0], and no metastases [M0]). Inclusion and exclusion criteria have been described previously.[20]

Patients were randomized 1:1 to receive either oral anastrozole 1 mg daily with a tamoxifen placebo or oral tamoxifen 20 mg daily with an anastrozole placebo. Both treatment groups received goserelin 3.6 mg as a subcutaneous injection every 28 days. Treatment continued for 24 weeks before surgery or until patients met any criterion for discontinuation.

The primary study endpoint was the best overall tumor response during the 24-week neoadjuvant treatment period. Secondary endpoints included histopathologic response, changes in estrone (E_1) and estradiol (E_2) serum and breast tumor tissue concentrations, changes in Ki-67 expression, and tolerability. For this exploratory analysis, we assessed correlations between Ki-67 expression and tumor response, ER status, PgR status, or histopathologic response.

The protocol was approved by an institutional review board at all study sites, and all enrolled patients provided written informed consent. The study (National Clinical Trials identifier NCT00605267) was conducted in accordance with the Declaration of Helsinki and good clinical practice, the applicable local regulatory requirements, and the AstraZeneca policy on Bioethics.

Assessments

Tumor measurements were performed using caliper measurements, ultrasound, or MRI or CT studies. The

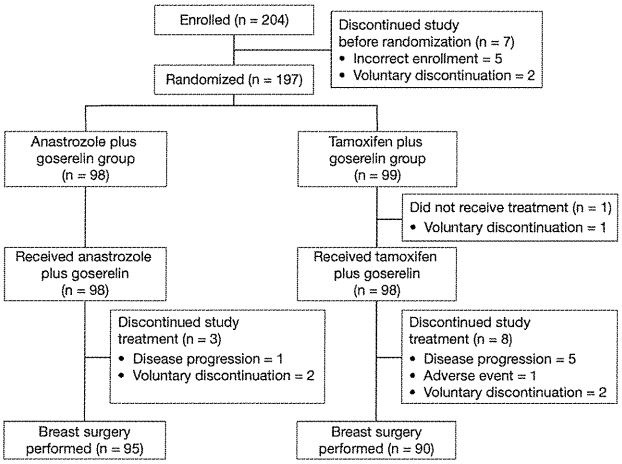

Figure 1. This is a CONSORT (Consolidated Standards of Reporting Trials) diagram of the current study.

primary analysis indicated that the best overall tumor response for anastrozole versus tamoxifen was consistent, regardless of the measurement method used.[20] We present tumor response data from the MRI or CT measurements at day 0 and at 24 weeks. The objective tumor response was assessed according to modified Response Evaluation Criteria in Solid Tumors (RECIST).[21]

The status of Ki-67, ER, and PgR was determined using histopathologic core-needle biopsy specimens that were collected at baseline and at surgery. Tissue sections were fixed in formalin and stored at room temperature before immunohistochemical staining. Ki-67 expression was determined by staining sections with an anti-MIB-1 antibody at a central laboratory (SRL Inc., Tokyo, Japan) for assessment by a central review board. For all slides, photomicrographs were taken from 3 to 5 hotspots at ×20 magnification using light microscopy. Two pathologists independently assessed the photomicrographs, and the Ki-67 index was calculated as the ratio of Ki-67-positive cancer cells from a total of 1000 cancer cells. ER-positive status and PgR-positive status at baseline were defined as ≥10% staining of cancer cell nuclei determined by a pathologist at each individual study site (nuclei were assessed using mouse monoclonal antibody clones 6F11 and 16, respectively). Staining for ER and PgR also was assessed in parallel using Allred scores by the Central Pathologist Review Committee.[22] An Allred score (the proportion score plus the intensity score) of ≥3 defined ER or PgR positivity, a score from ≥3 to <7 indicated medium expression, and a score of ≥7 indicated rich expression.

Histopathologic effects were assessed by comparing histopathologic samples that were obtained at baseline and at surgery. For the assessment of histopathologic response, the following categories were used: grade 0 indicated no response; grade 1a, marked change in <1 of 3 cancer cells; grade 1b, marked changes in ≥1 of 3 but <2 of 3 cancer cells; grade 2, marked changes in ≥2 of 3 cancer cells; and grade 3, necrosis or disappearance of all cancer cells and replacement of all cancer cells by granuloma-like and/or fibrous tissue. The histopathologic response was defined as the proportion of patients whose tumors were classified as grade 1b, 2, or 3.[23,24]

Post hoc subset analyses were used to determine correlations between the baseline Ki-67 index (≥20% vs <20%) and the best overall tumor response. The percentage change in the Ki-67 index for responders (patients whose best overall tumor response was a complete or partial response) versus nonresponders (patients whose best overall tumor response was stable or progressive disease) also was compared. Correlations between the baseline Ki-67 index and the histopathologic response at week 24 also were evaluated, and we used post hoc analyses to investigate correlations between changes in the Ki-67 index from baseline to week 24 and ER or PgR status at baseline. Positive ER and PgR status (Allred score ≥3) also was assessed at baseline and at week 24. Preoperative Endocrine Prognostic Index (PEPI) scores, which were calculated post hoc as the sum of risk points weighted by the size of the hazard ratio for tumor size, pathologic lymph node status, ER status, and Ki-67 expression for both recurrence-free and breast cancer-specific survival, were determined for each patient at surgery according to the methods described by Ellis and colleagues.[25]

Statistical Analysis

The sample size calculation and the main statistical analyses have been described previously.[20] All randomized patients were included in the intent-to-treat analysis set.

In a post hoc exploratory analysis, chi-square tests were performed to compare the best overall tumor response at week 24 between baseline Ki-67 index categories (≥20% vs <20%) within each treatment group and between treatment groups within each baseline Ki-67 index category. A chi-square test also was used to compare the histopathologic response at 24 weeks between the baseline Ki-67 index categories within each treatment group. All tests were made at the nominal 2-sided significance level of .05.

RESULTS

Patients

In total, 197 patients were randomized to receive either anastrozole plus goserelin (n = 98) or tamoxifen plus goserelin (n = 99) (Fig. 1). Patient demographics and

TABLE 1. Patient Demographics and Baseline Tumor Characteristics

Characteristic	No. of Patients (%)	
	Anastrozole Plus Goserelin	Tamoxifen Plus Goserelin
No. of patients	98	99
Age: Median [range]	44 [28 54]	44 [30 53]
Body mass index: Mean±SD, kg/m^2	22.2±3.5	22.1±3.3
Histology type		
Infiltrating ductal carcinoma	87 (88.8)	91 (91.9)
Infiltrating lobular carcinoma	3 (3.1)	3 (3)
Other[a]	8 (8.2)	5 (5.1)
Tumor grade		
1	42 (42.9)	48 (48.5)
2	36 (36.7)	26 (26.3)
3	4 (4.1)	14 (14.1)
Not assessable	1 (1)	0 (0)
Not done	15 (15.3)	11 (11.1)
Hormone receptor status		
ER positive	98 (100)	99 (100)
PgR positive	93 (94.9)	87 (87.9)
HER2 negative	98 (100)	99 (100)

Abbreviations: ER, estrogen receptor; HER2, human epidermal growth factor receptor 2; PgR, progesterone receptor; SD, standard deviation.
[a] Other included adenocarcinoma (n 3).

baseline characteristics generally were well balanced between the treatment groups (Table 1). Paired samples for calculating changes in the Ki-67 index from baseline to week 24 were available for 89 patients in the anastrozole plus goserelin group and for 86 patients in the tamoxifen plus goserelin group.

Correlation of the Baseline Ki-67 Index and Best Overall Tumor Response

With a mean baseline Ki-67 index of 21.9% and 21.6% in the anastrozole and tamoxifen treatment groups, respectively, we used post hoc subset analyses to compare patients according to their baseline Ki-67 index (≥20 vs <20%). For anastrozole versus tamoxifen, best overall tumor response from baseline to week 24 was better with anastrozole plus goserelin versus tamoxifen plus goserelin both in patients who had a baseline Ki-67 index ≥20% (73.2% vs 44.8%; $P = .002$) and in patients who had a baseline Ki-67 index <20% (52.5% vs 29%; $P = .035$) (Fig. 2A).

Within the treatment groups, the best overall tumor response from baseline to 24 weeks, as measured by MRI or CT, was significantly better with anastrozole plus goserelin for patients who had a baseline Ki-67 index ≥20% than for those who had a baseline Ki-67 index <20% (73.2% vs 52.5%; $P = .036$). Among patients in the tamoxifen plus goserelin group, the best overall tumor response was 44.8% for patients who had a baseline Ki-67 index ≥20% and 29% for those who had a baseline Ki-67 index <20% ($P = .118$) (Fig. 2A).

Correlation of the Baseline Ki-67 Index and Histopathologic Response

There was no significant difference in the histopathologic response between patients who had a baseline Ki-67 index ≥20% versus patients who had a baseline Ki-67 index <20% in either treatment group (Fig. 2B).

Correlation of Change in the Ki-67 Index and Responders/Nonresponders

A waterfall plot of changes in the Ki-67 index for individual patients, illustrated according to responders or nonresponders, is provided in Figure 3. There was no apparent relation between a change in Ki-67 expression from baseline to week 24 for responders and nonresponders in either treatment group.

Correlation of the Baseline Ki-67 Index and Estrogen Receptor or Progesterone Receptor Status

In both treatment groups, positive ER status, as determined by the Allred score, was observed in 100% of patients at baseline and at week 24, and >90% of patients in both treatment groups were ER rich (baseline Allred score, ≥7). Therefore, it was not possible to determine any potential relation between the baseline ER Allred score and the percentage change in Ki-67 expression from baseline to week 24 in either treatment group.

In the anastrozole plus goserelin group, 98.9% of patients were positive for PgR expression at baseline, and 34.4% were positive for PgR expression at week 24. The percentage of patients with positive PgR status was not altered from baseline (91.9%) to week 24 (89.5%) in the tamoxifen plus goserelin group (Fig. 4A). In both treatment groups, the mean decrease in the Ki-67 index was greater in patients who had a baseline PgR Allred score ≥7 (anastrozole group, −88.8%; tamoxifen group, −67.6%), compared with patients who had a baseline PgR Allred score <7 (anastrozole group, −74.1%; tamoxifen group, −32.8%) (Fig. 4B).

Preoperative Endocrine Prognostic Index Score

In the anastrozole treatment group, 33.3% of patients had a PEPI score of 0 compared with 11.4% in the tamoxifen group. Fewer patients (21.4%) had a PEPI score ≥4 in the anastrozole group compared with patients in the tamoxifen group (36.7%; $P = .002$) (Table 2).

DISCUSSION

In this exploratory analysis, we investigated changes in Ki-67 expression among patients from the STAGE study, a

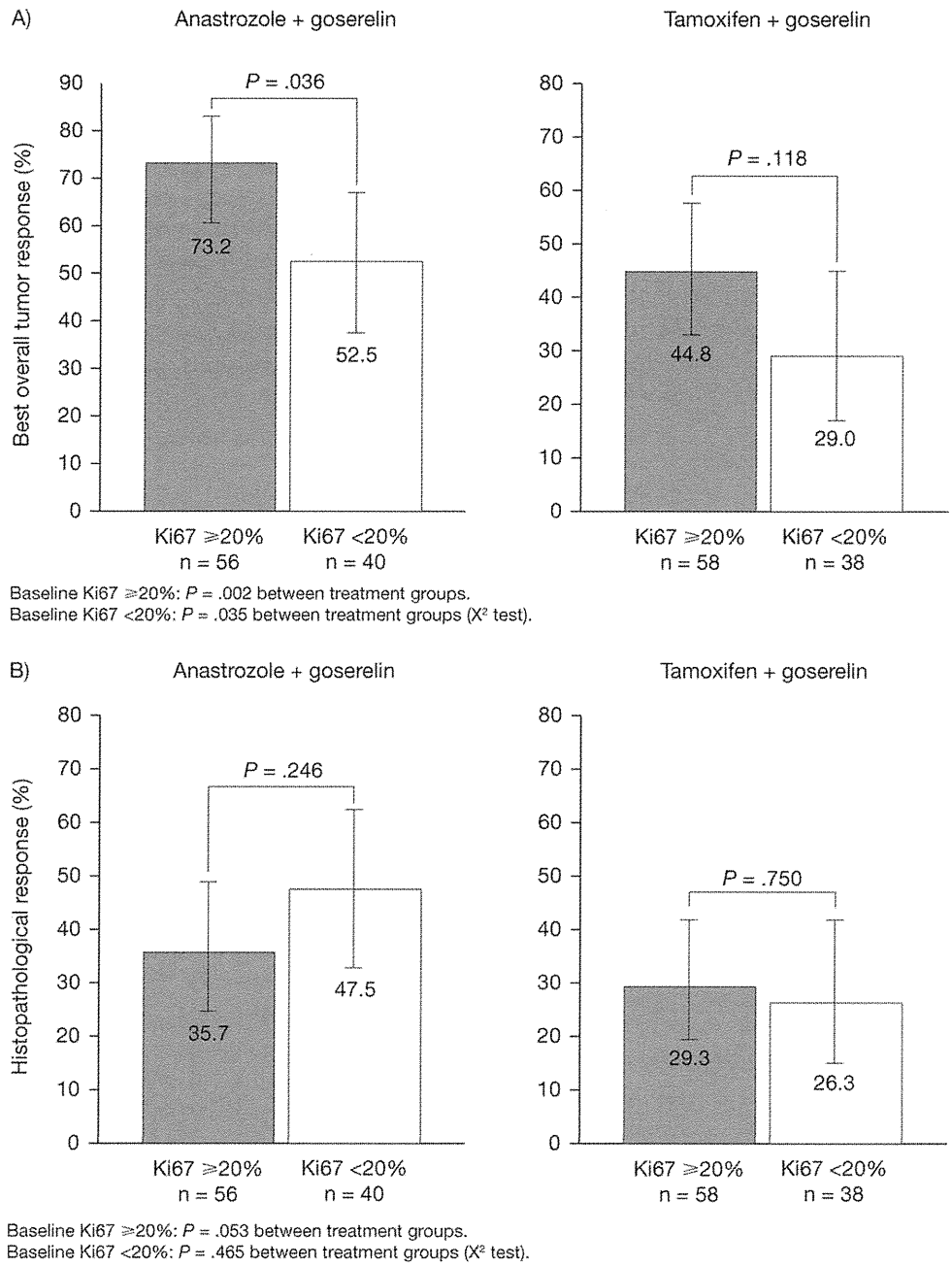

Figure 2. These charts illustrate the baseline Ki-67 index (≥20% vs <20%) according to (A) the best overall tumor response and (B) the histopathologic response at 24 weeks. Magnetic resonance imaging or computed tomography was used to measure responses. The best tumor response was defined a complete or partial response during the 24-week treatment period.

phase 3 randomized trial that compared tumor response for anastrozole plus goserelin versus response tamoxifen plus goserelin during 24 weeks of neoadjuvant treatment in premenopausal women with ER-positive breast cancer. The primary analysis indicated that the reduction in the Ki-67 index for patients who received goserelin was greater with anastrozole coadministration compared with tamoxifen, suggesting a greater inhibitory effect on tumor cell proliferation with this treatment combination.[20] Given the reported clinical prognostic value of Ki-67 expression after short-term neoadjuvant endocrine therapy for breast cancer,[19] this is in concordance with our finding that anastrozole combined with goserelin demonstrates a superior best overall tumor response compared with tamoxifen plus goserelin. Although Ki-67 is perceived as a reliable predictive endpoint, the outcomes of

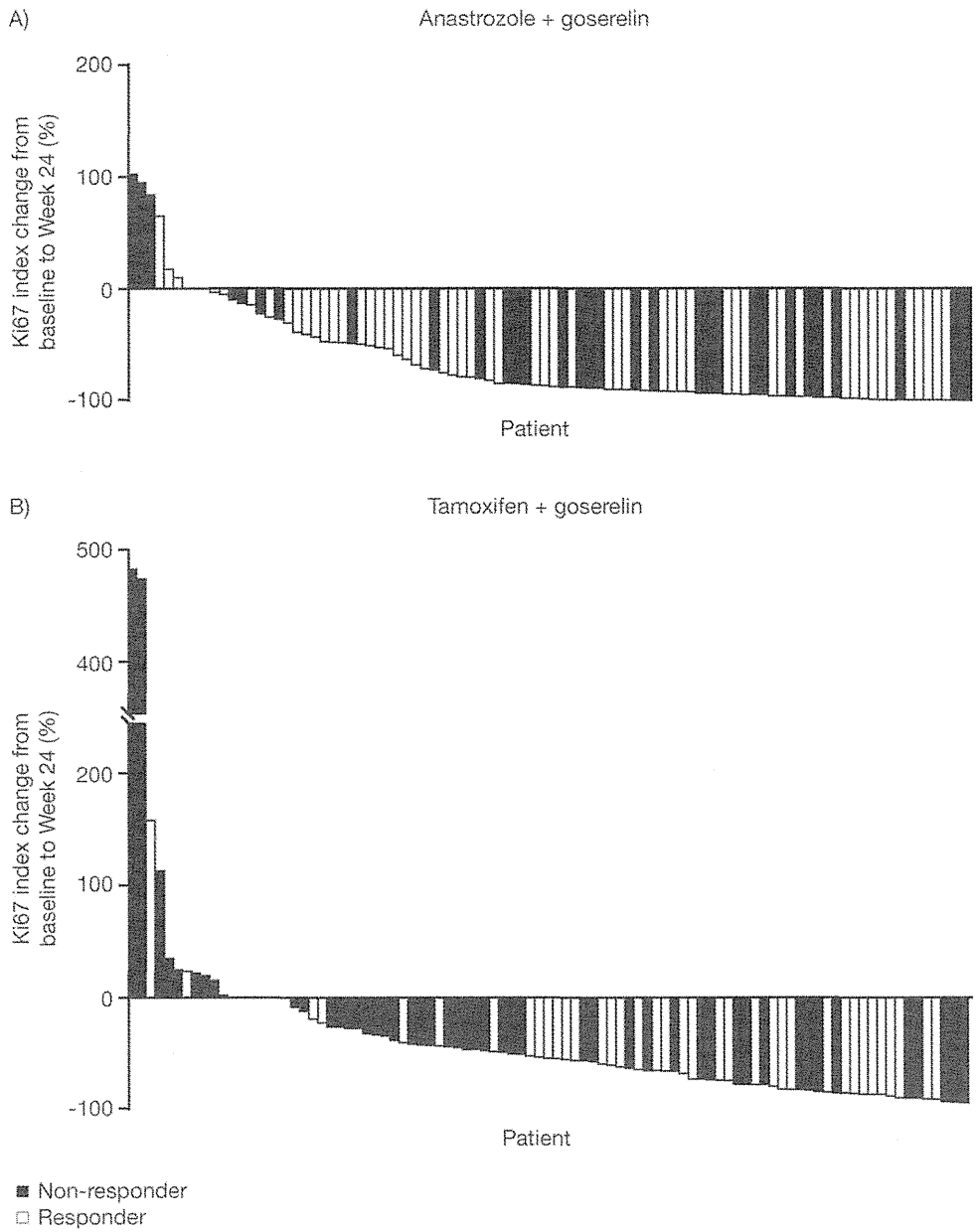

Figure 3. This is a waterfall plot of reductions in nuclear antigen Ki-67 levels in (A) the anastrozole plus goserelin treatment group and (B) the tamoxifen plus goserelin treatment group. Magnetic resonance imaging or computed tomography was used to measure responses. Responders were defined as those patients who had a complete or partial response during the 24-week treatment period.

the parallel adjuvant trial by the Austrian Breast and Colorectal Cancer Study Group (ABCSG) did not reflect outcomes related to the Ki-67 changes we observed: Results from the ABCSG-12 study indicated that there was no difference in disease-free survival between patients who received anastrozole versus tamoxifen (hazard ratio, 1.08; 95% CI, 0.81-1.44; $P = .591$).[26] The reason for this difference is not clear, although there were differences in the baseline characteristics of patients in each study: the STAGE study assessed a more hormone-dependent phenotype of tumor (ER-positive/HER2-negative in the STAGE study vs ER-positive/HER2-negative and ER-positive/HER2-positive in the ABCSG-12 trial), and the proportion of women with a body mass index >25 kg/m^2 was lower in the STAGE study (17% vs 33%). The ABCSG-12 group did not assess Ki-67 levels. It is also interesting to note that, as recently pointed out by Goncalves et al,[27] in our study, serum estradiol suppression

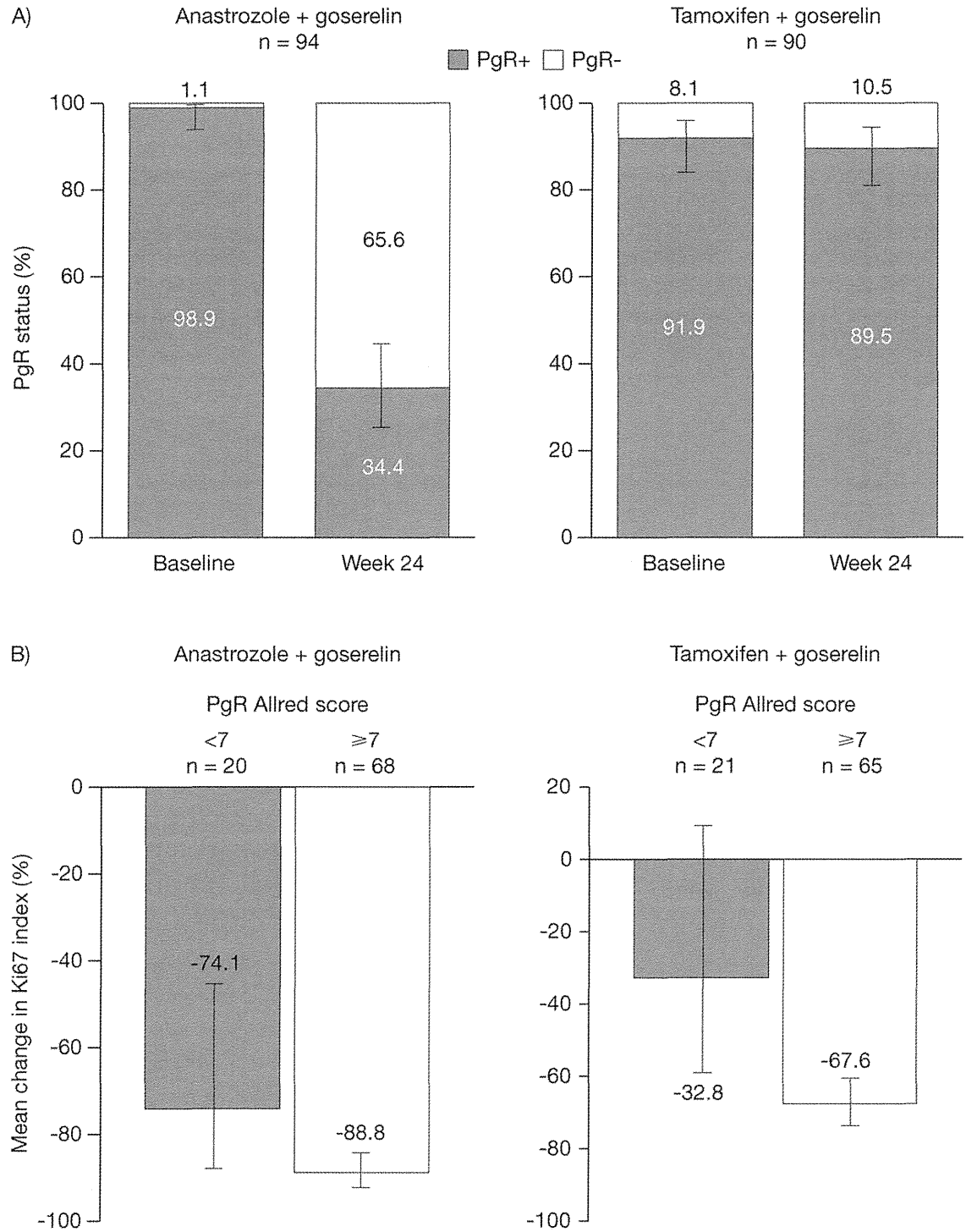

Figure 4. (A) Progesterone receptor status is illustrated at baseline and at 24 weeks. (B) Changes in the Ki-67 index and the baseline PgR Allred score are illustrated. PgR-positive (PgR+) indicates an Allred score >3; PgR-negative (PgR), an Allred score <2.

appeared to decrease at week 24 compared with week 4, although the suppression was not statistically significant. This suggests the possibility of a gradual tachyphylaxis of the estrogen-suppressing effects of combined goserelin and anastrozole treatment, which potentially may explain the difference in outcomes between the ABCSG-12 and STAGE studies. However, further investigations would be required to confirm this.

TABLE 2. Preoperative Endocrine Prognostic Index Score

Treatment Group	No. of Patients	PEPI Score: No. of Patients (%)		
		0	1-3	≥4
Anastrozole plus goserelin	84	28 (33.3)	38 (45.2)	18 (21.4)
Tamoxifen plus goserelin	79	9 (11.4)	41 (51.9)	29 (36.7)
P for anastrozole vs tamoxifen				.002

Abbreviation: PEPI, Preoperative Endocrine Prognostic Index.
[a]*P* values were determined using the chi square test.

In the current study, the best overall tumor response was superior with anastrozole compared with tamoxifen, irrespective of the baseline Ki-67 index. Within the anastrozole treatment group, we observed that the best overall tumor response was significantly better in patients who had a baseline Ki-67 index ≥20% versus patients who had a baseline Ki-67 index <20%. However, in the anastrozole group, we observed a numerically lower histopathologic response in patients who had a baseline Ki-67 index ≥20% compared with those who had a baseline Ki-67 index <20%. It was reported previously that baseline Ki-67 expression was not associated with outcome after neoadjuvant endocrine treatment (including anastrozole, letrozole, and tamoxifen) in ER-positive, postmenopausal women who had breast cancer.[19,25]

There was no apparent relation between a reduction in the Ki-67 index for responders and nonresponders in either treatment group. Although there tended to be more nonresponders among patients in the tamoxifen group who had less of a reduction in the Ki-67 index, the Spearman rank-correlation between the percentage change in the Ki-67 index and the best percentage change in greatest tumor dimension for the tamoxifen group was a modest 0.314. This observation is essentially consistent with what was reported previously by Dowsett et al, who conducted a similar analysis of postmenopausal patients who received neoadjuvant tamoxifen, anastrozole, and the tamoxifen/anastrozole combination.[28] This variation in the Ki-67 index change between responders and nonresponders indicates that the mechanism of estrogen-dependent growth is heterogeneous among breast tumors. Tumor growth is determined by a balance between cell proliferation and apoptosis. Stimulation of cell proliferation by estrogen may be dominantly implicated in tumor growth in some tumors, whereas inhibition of apoptosis by estrogen may be dominantly implicated in other tumors. Thus, a responder does not necessarily have a greater reduction in the Ki-67 index compared with a nonresponder if apoptosis is induced more strongly in the former than the latter after treatment.

In the neoadjuvant setting, endocrine therapy has demonstrated greater (or equivalent) efficacy in postmenopausal women with a lower Ki-67 index.[29,30] In contrast, in our study, both anastrozole and tamoxifen produced greater response rates in premenopausal women with a higher Ki-67 index. It is therefore possible that the main pathways of proliferative stimulation (and the effectiveness of endocrine treatments) may differ between premenopausal and postmenopausal women with ER-positive breast cancer, according to their level of Ki-67 expression. In general, high Ki-67 expression is traditionally is believed to offer a poor prognosis and is predictive of response to chemotherapy regimens.[31] However, our results suggest that endocrine therapy has at least comparable effectiveness for premenopausal patients with ER-positive breast cancer who have a high Ki-67 index.

No correlation could be determined between a change in the Ki-67 index and baseline ER status in either treatment group. However, the number of patients who were identified as PgR-positive decreased at week 24 in the anastrozole treatment group, an effect that was not observed in the patients who received tamoxifen plus goserelin. PgR expression also was reduced under neoadjuvant AI treatment for breast cancer in the ABCSG 17 study, although it remains to be determined whether the down-regulation of PgR may be used as a marker of clinical efficacy.[32] In our study, the reason why the positive rate of PgR was reduced in the anastrozole plus goserelin arm compared with the tamoxifen plus goserelin arm is most likely because of the estrogenic action of tamoxifen, which would induce PgR expression.

Although there may be a potential correlation between a reduction in Ki-67 and the baseline PgR Allred score in patients who receive anastrozole plus goserelin versus tamoxifen plus goserelin, further analyses will be required to determine whether a Ki-67 reduction in patients with high baseline PgR expression translates into a clinical benefit.

After treatment with anastrozole, a lower proportion of patients had a PEPI score ≥4 (indicating a high risk of

recurrence) compared with the tamoxifen treatment group. The PEPI model has been validated previously and has indicated significant differences in recurrence-free survival in the adjuvant setting between 3 PEPI risk groups (PEPI risk scores of 0, 1-3, and ≥4), with a PEPI score of 0 indicating a very low risk of relapse.[25] Data from the adjuvant treatment setting will provide added knowledge for the individualization of future adjuvant treatments after neoadjuvant therapy for breast cancer.

Currently, very little is known about the prognostic effect of Ki-67 in premenopausal women. However, in 1 recent study, the prognostic significance of Ki-67 was investigated in women with ER-positive breast cancer who had received short-term presurgical tamoxifen, and Decensi and colleagues reported that the Ki-67 response was a good predictor of recurrence-free survival and overall survival.[33]

To our knowledge, this is the first randomized study to investigate the potential of Ki-67 as a clinical biomarker for AI efficacy in premenopausal women with ER-positive breast cancer. It has been demonstrated that a reduction in Ki-67 expression as a result of neoadjuvant AI treatment can be a potentially useful marker of improved surgical outcomes in postmenopausal women with ER-positive breast cancer, and such a reduction has been identified as predictive of favorable outcomes in the adjuvant treatment period.[34] A reduction in Ki-67 expression during neoadjuvant treatment reportedly was greater with anastrozole versus tamoxifen in postmenopausal women who had ER-positive breast cancer,[18] and a parallel result also was observed in the corresponding adjuvant trial, in which recurrence-free survival also was greater for those who received anastrozole.[8] Yet another similar result was observed for letrozole, in which a greater Ki-67 reduction was observed compared with tamoxifen in the neoadjuvant setting.[35] Greater clinical effectiveness also was observed for letrozole in the neoadjuvant setting, both in terms of the objective response rate and the rate of breast-conserving surgery.[36]

In conclusion, tumor response was greater with anastrozole compared with tamoxifen, regardless of the baseline Ki-67 index, in premenopausal women who received goserelin as neoadjuvant therapy for ER-positive, early stage breast cancer. The current results indicate that endocrine therapy may offer a more tolerable treatment option than cytotoxic chemotherapy as neoadjuvant treatment for these patients, and further studies of the anastrozole plus goserelin treatment combination in this setting are warranted.

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CONFLICT OF INTEREST DISCLOSURES

Dr. Iwase has received honoraria from AstraZeneca and research funding from AstraZeneca; Chugai Pharmaceutical Company, Ltd.; Novartis; and Takeda. Mr. Hayashi is an employee and holds stock ownership with AstraZeneca. Dr. Noguchi has received honoraria and research funding from and has acted in a consultant or in an advisory role for AstraZeneca.

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ORIGINAL ARTICLE

A multicenter, phase II study of epirubicin/cyclophosphamide followed by docetaxel and concurrent trastuzumab as primary systemic therapy for HER-2 positive advanced breast cancer (the HER2NAT study)

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Abstract
Background The outcome in patients with human epidermal growth factor receptor-2 (HER-2)-positive locally advanced breast cancer may be improved by integrating trastuzumab with primary systemic therapy (PST).
Methods The efficacy and safety of PST comprising EC (epirubicin 90 mg/m^2 and cyclophosphamide 600 mg/m^2, four cycles every 3 weeks) followed by docetaxel (75 mg/m^2, four cycles every 3 weeks) and concurrent trastuzumab (loading dose 4 mg/kg followed by 2 mg/kg, 12 cycles every week) was investigated in a multicenter, prospective, phase II study in patients with HER-2-positive stage IIIB/IIIC/IV breast cancer. The primary endpoint was pathologic complete response (pCR) including the tumor intraductal component confirmed by central pathologic review.
Results In total, 38 patients were enrolled (stage IIIB, 63.2 %; IIIC, 23.7 %; IV, 13.2 %; estrogen receptor- and/or progesterone receptor-positive, 47.4 %). The pCR rate was 16.2 % in the primary tumor (six of 37 patients in the Full Analysis Set) and 56.8 % (21/37) in the ipsilateral

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axillary lymph nodes. Treatment was given according to protocol in 28 of 37 patients; six of 28 in the Per-Protocol Set achieved pCR (21.4 %). The clinical response rate was 67.6 % (25/37 patients; complete response, 13.5 %; partial response, 54.1 %). No patients developed congestive heart failure; however, three patients had a non-symptomatic decrease of >10 % of left ventricular ejection fraction.
Conclusions PST including concurrent use of trastuzumab combined with docetaxel is effective and well-tolerated in HER-2-positive advanced breast cancer patients, including those patients requiring mastectomy for local control.

Keywords Primary systemic therapy · HER-2 · Trastuzumab · Advanced breast cancer

Introduction

The clinical outcome for patients with locally advanced or metastatic breast cancer (stages IIIB, IIIC, IV) is poor compared with those presenting with operable disease [1]. Anthracycline taxane-based chemotherapy is the mainstay of treatment, but the therapeutic strategy needs to be tailored to individual patient characteristics. Neoadjuvant or primary systemic therapy (PST) is accepted treatment for patients with locally advanced breast cancer, but there is no standard regimen [2]. The advantages of PST in this setting include earlier treatment of subclinical distant micrometastases, downstaging of the primary tumor to facilitate surgery, and providing an opportunity to assess response to treatment in vivo [1]. Highly active PST can result in the disappearance of all invasive and non-invasive tumors [pathologic complete response (pCR)]. Since achievement of pCR correlates with improved survival in patients with operable breast cancer, pCR is accepted as a valid endpoint for trials of PST [3, 4].

The human epidermal growth factor receptor-2 (HER-2), which is amplified or overexpressed in approximately 25 % of breast cancers, is associated with aggressive disease and a poor prognosis [5]. Although it is an adverse prognostic factor, HER-2 has also emerged as a favorable predictive factor following the development of a specific treatment targeting this molecular abnormality [6]. Trastuzumab (Herceptin®, Roche, Basel, Switzerland) is a recombinant humanized monoclonal antibody targeting HER-2 that has demonstrated clinical efficacy as monotherapy [7 10] and in combination with chemotherapy in patients with HER-2-positive metastatic [10 12] and early breast cancer [13, 14]. Pivotal trials have shown that trastuzumab is well tolerated, although the potential for cardiotoxicity means that caution is required when considering concurrent or sequential use in patients receiving anthracycline-based regimens [15].

Early clinical studies in patients with HER-2-positive locally advanced breast cancer have shown that the combination of trastuzumab with taxane-based PST achieves a high pCR rate [16]. However, since anthracycline taxane regimens are the most frequently prescribed treatments in patients with operable breast cancer [2] and it is not known whether the efficacy of non-anthracycline regimens is equivalent, there is a compelling argument to investigate the addition of trastuzumab to anthracycline taxane PST. Although concurrent use is not recommended, it is feasible to administer trastuzumab sequentially after epirubicin, which is less cardiotoxic than doxorubicin, provided that a sub-cardiotoxic cumulative dose of anthracycline has been received [17].

A multicenter, prospective phase II study was therefore conducted in Japan to evaluate the efficacy of trastuzumab added to standard anthracycline taxane-based PST for patients with locally advanced or metastatic HER-2-positive breast cancer. The regimen was designed to achieve a high pCR rate.

Patients and methods

The primary objective of this phase II study was to determine the pCR rate of trastuzumab-containing PST in patients with HER-2-positive locally advanced or metastatic breast cancer. Secondary objectives of the study included clinical response rate (cRR), pCR in ipsilateral axillary nodes, and safety and toxicity of the PST regimen.

Patient selection

Patients eligible for the study had a histologically confirmed diagnosis of primary breast cancer and locally advanced disease according to tumor node metastasis staging (T4 or N3 or M1, clinical stages IIIB, IIIC, or IV). Patients with inflammatory breast cancer were excluded. Only patients with HER-2-positive tumors determined by immunohistochemistry (IHC 3+) and/or fluorescence in-situ hybridization (FISH+) were eligible for inclusion. HER-2 testing was performed locally according to the protocol used at the participating center. Other key eligibility criteria included age $\geq$20 years and $\leq$65 years, performance status 0 1, left ventricular ejection fraction (LVEF) measured by echocardiography or multi-gated acquisition (MUGA) scan $\geq$55 %, white blood count $\leq$10,000/mm^3, absolute neutrophil count $\geq$2,000/mm^3, platelet count $\geq$100,000/mm^3, hemoglobin $\geq$9.5 g/dl, alanine aminotransferase (ALT) and aspartate aminotransferase (AST) $\leq$60 IU/l, bilirubin $\leq$1.5 mg/dl, and creatinine $\leq$1.5 mg/dl. The protocol was reviewed and approved by the institutional review board of each participating center. All patients provided written informed consent.

Treatment

Eligible patients were scheduled to receive PST comprising four cycles of EC (intravenous epirubicin 90 mg/m^2 and intravenous cyclophosphamide 600 mg/m^2, administered on day 1 every 3 weeks) followed by four cycles of docetaxel (75 mg/m^2 intravenously, on day 1 every 3 weeks) and 12 cycles of concurrent trastuzumab (loading dose 4 mg/kg intravenously followed by 2 mg/kg, on day 1 every week).

Trastuzumab doses were modified in patients developing a decline in LVEF or congestive heart failure (CHF) according to the protocol used in the Herceptin Adjuvant (HERA) trial [18]. Trastuzumab was discontinued if the patient experienced symptomatic CHF and an LVEF of $\leq$45 or <50 % with an absolute reduction of $\geq$10 % from baseline. Trastuzumab was also discontinued in asymptomatic patients if LVEF did not return to a level above the criteria for withholding treatment after therapy was stopped for 3 weeks.

After completion of PST, patients were scheduled to receive surgery. Patients with stage IV disease could elect to receive salvage mastectomy for local control. Adjuvant trastuzumab therapy was administered after surgery. Patients could receive one of two trastuzumab regimens at the physician's discretion: loading dose 4 mg/kg followed by 2 mg/kg every week for 40 cycles or loading dose 8 mg/kg followed by 6 mg/kg every 3 weeks for 14 cycles. Patients with hormone-receptor positive disease also received adjuvant endocrine therapy for 5 years: premenopausal women received tamoxifen for 5 years, while postmenopausal women received tamoxifen, letrozole or anastrozole. The treatment protocol is summarized in Fig. 1.

Assessment of endpoints

The primary endpoint of the study was pCR, defined as the absence of invasive components of ductal or lobular carcinoma in the breast with pathologic review confirming necrosis and/or disappearance of all tumor cells, and/or the replacement of cancer cells by granulation and/or fibrosis [19]. A central pathology review board confirmed all pCRs. Near pCR was defined as grade 2b, the absence of invasive component of ductal or lobular carcinoma in the breast with pathologic findings showing extremely high-grade tumor and marked changes approaching a complete response with only a few cancer cells remaining [19]. Near pCR was determined by pathologists at the local institutions.

The secondary endpoints of the study were cRR, pN0 in the ipsilateral axillary lymph nodes, and safety and toxicity. Clinical responses in the primary tumor were assessed by computed tomography (CT), ultrasonography (US) and/or by palpation. CT, US, or magnetic resonance imaging (MRI) were used to assess clinical responses in axillary lymph node tumors. Clinical responses were defined according to Response Evaluation Criteria In Solid Tumors [20].

Adverse events were reported and graded according to Common Terminology Criteria for Adverse Events v3.0 [21]. Cardiac function was monitored regularly using echocardiography or MUGA scans. Cardiac scans and electrocardiograms were performed at baseline, and after 12 weeks (four courses) and 25 weeks (completion of therapy) in the neoadjuvant setting, or at 3 months, 6 months and at the end of therapy (40 weeks) in the adjuvant setting. CHF and LVEF dysfunction were defined as described in the HERA trial [18].

Statistics

The planned study sample size was 40 patients based on achieving a pCR of 3 % in the null hypothesis or 18 % in the alternative hypothesis ($\alpha = 0.05$; $\beta = 0.1$), assuming a rate of about 30 % of patients excluded from the analysis set [16, 22, 23].

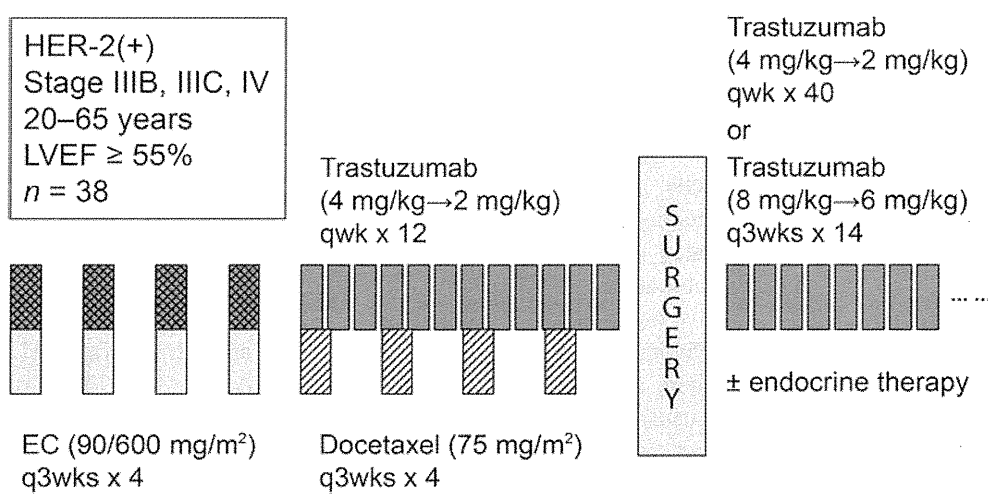

Fig. 1 Study design and treatment protocol

All registered patients were included in the analysis for safety and toxicity. The Full Analysis Set (FAS) comprised subjects meeting the inclusion criteria specified in the protocol at the data center and treated with at least one dose of protocol-specified therapy. Subjects from the FAS who underwent protocol-specified surgical procedures and did not have any serious deviations from the treatment protocol were included in the Per-Protocol Set (PPS). Protocol deviations resulting in exclusion from the PPS included failure to undergo surgery and administration of <80 % of the total dose of treatment. The data adjusted for FAS and PPS were used for the analyses of efficacy.

Subgroup analyses were performed using Fisher's exact test to determine associations between pCR rate and hormone receptor status [estrogen receptor/progesterone receptor (ER/PgR) positive or negative] or tumor size (T4 vs. T1, 2, or 3). Fisher's exact test was also used to determine whether CT, US, or MRI were more effective than palpation in predicting pCR based on clinical complete response (cCR).

Results

The study was performed in 11 centers in Japan between January 2006 and December 2008. In total, 38 patients entered the study and were included in the safety analysis. The characteristics of all registered patients are shown in Table 1. One patient with HER-2-negative breast cancer was ineligible and therefore the FAS comprised 37 patients. Surgery was not performed in three patients and a further six patients received <80 % of the total planned treatment dose. Consequently, 28 patients were included in the PPS.

Table 1 Patient characteristics

Patient characteristic	*n* 38
Median age (range)	49.5 years (39 64)
Menopausal status (*n*)	
Pre	22
Post	16
Tumor size (*n*)	
T1	1
T2	6
T3	4
T4	27
Axillary lymph node status (*n*)	
N0	1
N1	14
N2	7
N3	16
Clinical stage (*n*)	
IIIB	24
IIIC	9
IV	5
ER/PgR status (*n*)	
ER and/or PgR positive	18
ER and PgR negative	20
HER 2 status (IHC3+ and/or FISH+; *n*)	
Positive	37
Negative	1[a]

ER estrogen receptor, *FISH* fluorescence in situ hybridization, *IHC* immunohistochemistry, *PgR* progesterone receptor

[a] Ineligible

[b] All patients were node positive

Dose delivery

The median dose delivered was equivalent to the planned total dose for all agents in the PST regimen (Table 2). The mean doses of chemotherapy delivered were >90 % for all regimens and the mean dose of trastuzumab delivered was 88 % of the planned total dose (Table 2).

Efficacy of PST in the primary tumor

The pCRs and cRRs achieved in the primary tumor are shown in Table 3. The pCR rate was six of 37 patients [16.2 %; 95 % confidence interval (CI) 6.2 32.0] in the FAS and six of 28 patients (21.4 %; 95 % CI 8.3 41.0) in the PPS (Fig. 2a). In addition, 10 patients in the FAS and nine in the PPS achieved Near pCR; therefore, the rates of pCR plus Near pCR were 43.2 and 53.6 %, respectively (Fig. 2a). The overall cRR was 25 of 37 patients (67.6 %) in the FAS and 24 of 28 patients (85.7 %) in the PPS. The cCR rates were 13.5 and 17.9 %, respectively. All patients achieving pCR in the primary tumor were shown to have pN0 status in the axillary lymph nodes.

The rate of pCR plus Near pCR was higher in patients with ER/PgR-negative disease than in those with ER/PgR-positive disease (55.0 vs. 29.4 %, respectively); however, there was no significant association between pCR rate and hormone-receptor status ($P = 0.1886$; Fig. 2b). The rates of pCR plus Near pCR were similar in patients with T1, 2, or 3 and those with T4 tumors (45.5 vs. 42.3 %, respectively), but the pCR rate was low in patients with T4 tumors (36.4 vs. 7.7 %, respectively; $P = 0.0515$; Fig. 2c).

Efficacy of PST in axillary lymph nodes

All patients were pathologically axillary lymph node positive prior to treatment even though one patient was clinically negative. Following PST, 21 of 37 patients (56.8 %) in the FAS had no lymph-node involvement

Table 2 Actual dose delivery

	Treatment			
	Epirubicin (dose in mg/m^2)	Cyclophosphamide (dose in mg/m^2)	Docetaxel (dose in mg/m^2)	Trastuzumab (dose in mg/kg)
Number of patients	38	38	36	37
Mean dose (range)	347 (90 377)	2,346 (600 2516)	273 (75 318)	23 (4 28)
Standard deviation	47	309	63	7
Median dose	360	2,408	300	26
Planned dose	360	2,400	300	26

Table 3 Efficacy of primary systemic therapy assessed in the primary tumor

Efficacy endpoint	Clinical outcome	FAS, *n* 37 (%)	PPS, *n* 28 (%)
Pathologic response rate (pCR)	pCR	6 (16.2)	6 (21.4)
	Near pCR	10 (27.0)	9 (32.1)
	pCR + Near pCR	16 (43.2)	15 (53.6)
Clinical response rate	Complete response	5 (13.5)	5 (17.9)
	Partial response	20 (54.1)	19 (67.9)
	Stable disease	3 (8.1)	2 (7.1)
	Progressive disease	2 (5.4)	1 (3.6)
	Not evaluable	7 (18.9)	1 (3.6)
	cRR	25 (67.6)	24 (85.7)

cRR clinical response rate (complete response and partial response), *FAS* Full Analysis Set, *PPS* Per Protocol Set

(pN0). Of these 21 patients, lymph-node positivity prior to PST was initially confirmed by biopsy in three, confirmed without biopsy in 14, and unclear in four patients. In the PPS, 20 of 28 patients (71.4 %) were pN0 following PST (lymph-node positivity confirmed by biopsy prior to PST in three patients, confirmed without biopsy in 13, and unclear in four).

Association between cCR and pCR

There was a significant association between cCR and pCR in the primary tumour when clinical response was assessed by palpation ($P = 0.0015$), but not by CT or US (Table 4a, b). Table 4c shows a significant association between cCR and pN0 when clinical response in axillary lymph-node tumors was assessed by CT, US, or MRI ($P = 0.0090$).

Safety

The trastuzumab-containing PST regimen used in this study was feasible and well tolerated. The predominant serious adverse events (grade 3/4) toxicity were hematologic (Table 5). More than 60 % of patients experienced grade 3 or 4 neutropenia, but the rate of febrile neutropenia was only 5.3 % (grade 3). The only other serious adverse event occurring at a frequency greater than 5 % (Table 5) was allergic reaction (grade 4 in 7.9 %: infusion reaction with trastuzumab in 2.6 % and allergic reaction with docetaxel in 5.3 %). No grade 3 edema associated with docetaxel was reported.

No cases of CHF were observed in the overall study population and no patients experienced a decrease in LVEF to <50 %. A decrease of >10 % in LVEF was reported in three patients (8.1 %). The decrease in LVEF prior to surgery was 10 % in one patient and 13 % in two patients. Trastuzumab was continued and all three patients underwent surgery.

Discussion

This study was designed to investigate whether addition of trastuzumab to a standard anthracycline taxane-based PST regimen could achieve a high pCR rate in patients with HER-2-positive locally advanced or metastatic breast cancer. The pCR rate was 16.2 % in the FAS and 21.4 % in the PPS. The findings of this study add to the weight of evidence supporting the addition of trastuzumab to anthracycline taxane-based PST for patients with HER-2-positive breast cancer [4, 18, 22, 24, 25], including those patients being treated in Japan.

In a pioneering study reported by Buzdar and colleagues [22], the addition of trastuzumab to paclitaxel-containing PST resulted in a significant improvement in the pCR rate from 25 to 66.7 % ($P = 0.02$). This study included patients with mainly T2 disease, and so focused on rather early stage tumors. In the large GeparQuattro study, which also

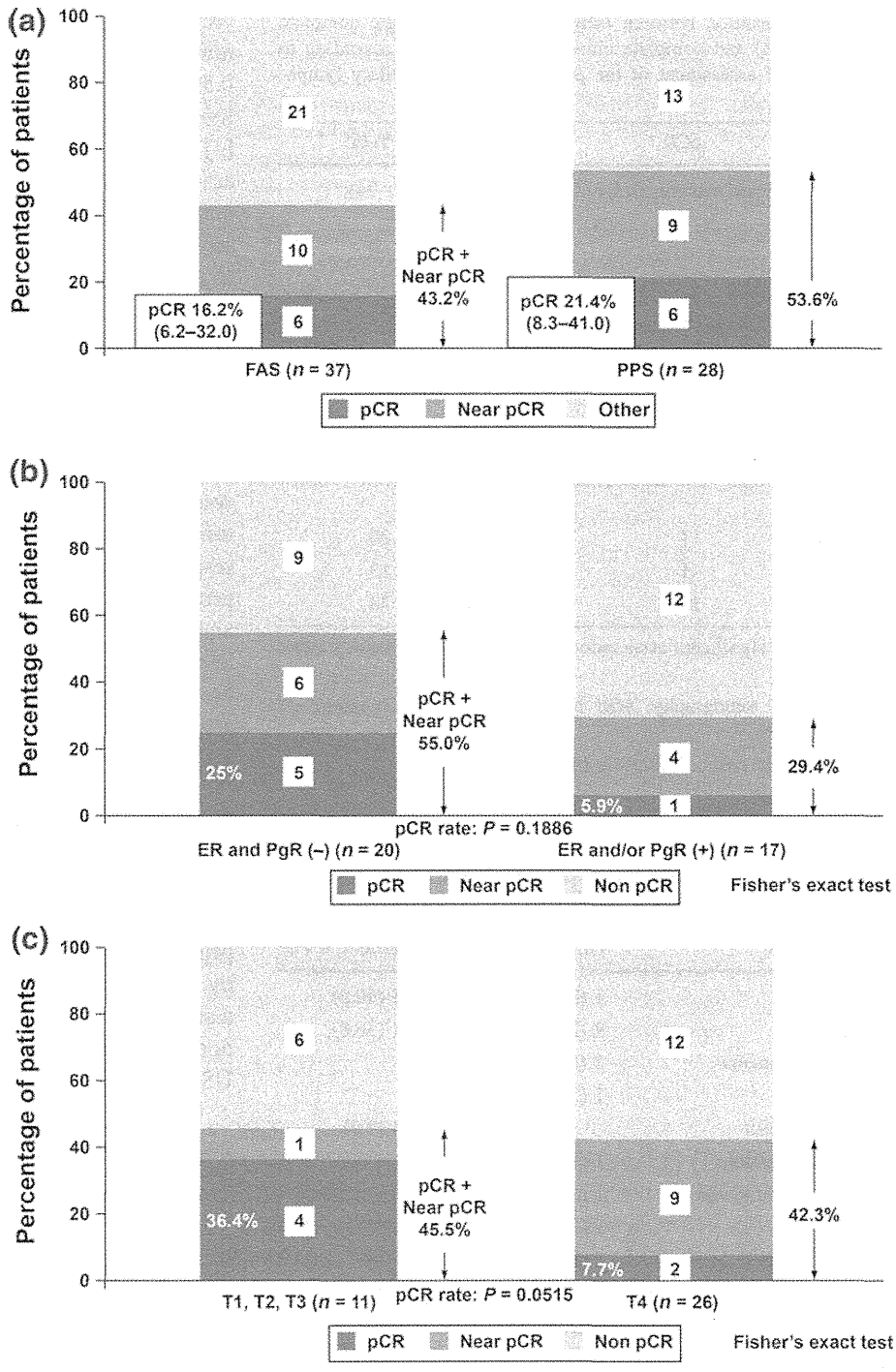

Fig. 2 Pathologic response rate (*pCR*) according to **a** FAS or PPS data set, **b** hormone receptor status (*n* 37), and **c** tumor size (T1 T3 vs. T4; *n* 37). *ER* estrogen receptor, *FAS* Full Analysis Set, *LVEF* left ventricular ejection fraction, *PPS* Per Protocol Set, *PgR* progesterone receptor

included mainly T2 patients, the docetaxel-containing PST led to a pCR rate of 31.7 % in the HER-2-positive group [17]. In the randomized NOAH trial, which included patients with later stage disease, similar to our study, 235 patients with HER-2-positive locally advanced breast cancer (100 T4 non-inflammatory, 63 inflammatory, and 72 N2 or ipsilateral node positive) were allocated to paclitaxel and doxorubicin-containing PST with or without trastuzumab [24]. Addition of trastuzumab significantly improved the rate of pCR from 22 to 43 % ($P = 0.0007$).

Comparing pCR rates between trials is difficult because of heterogeneous patient populations, different chemotherapy regimens, and variations in the assessment and definition of pCR. Nevertheless, it is instructive to use the published data as a benchmark to assess pCR rates reported in our study. Analysis of efficacy according to tumor size

Table 4 Association between achievement of pathologic complete response (pCR) and complete clinical response (cCR) according to the method of assessment of the primary tumor or axillary lymph node tumor

	cCR	Non cCR	Total
(a) Primary tumor assessment by CT, US, or MRI[a] (*n* 34); *P* 0.2053			
pCR	2	4	6
Non pCR	3	25	28
Total	5	29	34
(b) Primary tumor assessment by palpation (*n* 33); *P* 0.0015			
pCR	6	0	6
Non pCR	7	20	27
Total	13	20	33
(c) Axillary lymph node assessment by CT, US, or MRI (*n* 33); *P* 0.009			
pN0	11	9	20
Non pN0	1	12	13
Total	12	21	33

Probability of significant association determined using Fisher's exact test

CT computed tomography, *MRI* magnetic resonance imaging, *US* ultrasonography

[a] There was no case assessed by MRI

Table 5 Serious adverse events (CTCAE v3.0) reported in the overall study population (*n* 38)

Adverse event	Grade 3, *n* (%)	Grade 4, *n* (%)
Neutropenia	1 (2.6)	23 (60.5)
Leukopenia	9 (23.7)	14 (36.8)
Febrile neutropenia	2 (5.3)	
Anemia	1 (2.6)	
Allergic reaction		3 (7.9)
Rash/desquamation	1 (2.6)	
Rash/hand foot syndrome	1 (2.6)	
Anorexia	1 (2.6)	
Taste alteration	1 (2.6)	
AST elevation	1 (2.6)	
ALT elevation	1 (2.6)	
Dizziness	1 (2.6)	
Sensory neuropathy	1 (2.6)	
Joint pain	1 (2.6)	
Muscle pain	1 (2.6)	

ALT alanine aminotransferase, *AST* aspartate aminotransferase, *CTCAE* Common Terminology Criteria for Adverse Events

showed that the pCR rate among patients with T1 3 tumors was 36.4 %, which is similar to previous reports. Achievement of pCR is an important goal in neo-adjuvant treatment since it is a surrogate marker for better prognosis. In the NOAH trial, trastuzumab added to PST resulted in a significant improvement in event-free survival (hazard ratio 0.59; 95 % CI 0.38 0.90; $P = 0.013$) [24]. In addition, long-term follow-up of the study by Buzdar and colleagues [4] showed significantly improved disease-free survival among patients randomized to PST plus trastuzumab compared with chemotherapy alone. The pCR correlated with longer survival based on reports from the TECNO trial [25].

Differences in the pCR rate were evident when the efficacy data were analyzed according to patient characteristics. Consistent with data from the GeparQuattro study [17, 26], the probability of achieving pCR was higher in patients with hormone-receptor-negative disease. Although the pCR rate was highest in patients with T1 3 tumors, it was interesting to note that some patients with bulky tumors, including those with T4 or stage IV disease, did respond to the PST regimen used in this study. Better prognosis is expected in stage IV patients achieving pCR and surgery [27, 28]. Data have shown that pN0 is an excellent prognostic factor, even in patients with residual breast disease [29]. In the present study, the pathologic response rate in the axillary lymph nodes was 56.8 % in the FAS and 71.4 % in the PPS.

Achievement of pCR in the primary tumor and of pN0 in the axillary lymph nodes is a more relevant prognostic factor than clinical response in the setting of PST [30]. However, cCR may be predictive of pCR. In this study there was a significant association between cCR in the primary tumor and pCR when clinical assessment was done by palpation rather than CT or US. Furthermore, there was a significant association between cCR in the axillary lymph nodes and pN0 when clinical response was assessed by CT, US, or MRI.

Cardiotoxicity was identified as the major clinical problem in the upfront use of trastuzumab in combination with anthracycline-based regimens [11, 31]. In this study, the risk of cardiac toxicity was minimized by using epirubicin rather than doxorubicin and by sequential rather than concurrent administration of trastuzumab. Three patients experienced an asymptomatic decrease of >10 % in LVEF; however, no patient was unable to receive surgery because of cardiotoxicity and there were no cases of CHF reported. This observation supports the hypothesis that trastuzumab is associated with type II reversible cardiac dysfunction distinct from cardiotoxicity associated with anthracyclines [32]. Other investigators have used non-anthracycline-based regimens, including taxanes with or without platinum agents, as the foundation for trastuzumab-containing PST in patients with HER-2-positive breast cancer [17, 33 35]. It is not clear, however, if the omission of anthracycline from PST will adversely affect long-term outcome.

Our findings are in line with data from larger studies in patients with HER-2-positive operable or locally advanced breast cancer treated with trastuzumab in combination with anthracycline taxane PST [17, 24]. Although HER-2-positive T4 tumors showed a lower pCR rate than T1 T3 tumors, there was no difference in pathologic response rate including Near pCR between T4 and T1 T3 tumors. Strong PST is one of the treatment options for T4 tumors; it could improve the pCR rate of HER2-positive T4 tumors, especially in ER/PgR-negative and M0 patients. Strong PST is defined as high-dose, prolonged, dose-dense or combination of trastuzumab with anthracyclines.

Conclusion

For patients with HER-2-positive locally advanced and metastatic breast cancer, combining trastuzumab with an anthracycline taxane-based PST might achieve high pCR without significant toxicity.

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Conflict of interest The authors declare no conflicts of interest.

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SPECIAL FEATURE

Preoperative CT evaluation of intraductal spread of breast cancer and surgical treatment

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Abstract It is always a challenge to accurately determine the appropriate extent of resection in breast-conserving surgery (BCS), in order to reduce the need for re-excision, prevent local recurrence, and optimize cosmetic results. Detecting intraductal spread alone with high sensitivity may not be enough to realize safe BCS. Computed tomography carried out with the patient in the supine position accompanied by adequate marking is effective for preoperative determination of the optimum extent of BCS.

Keywords Breast cancer · CT · Breast-conserving surgery · Extent of surgery · Extensive intraductal component

Abbreviations

BCS	Breast-conserving surgery
CT	Computed tomography
EIC	Extensive intraductal component
HU	Hounsfield units
MD-CT	Multidetector-row computed tomography
MIP	Maximum intensity projection
MMG	Mammography
US	Ultrasonography

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Breast cancer diagnosis

Although computed tomography (CT) is not a primary modality for screening the breast or differentiating between malignant and benign breast lesions, some studies have reported that CT was able to reveal the primary tumor with high sensitivity [1]. Diagnostic criteria for breast cancer using CT include an irregular margin, irregular shape, and rim enhancement [2]. Spiculation was strongly suggestive of malignancy when detected incidentally by use of CT [3 5]. Irregular shape and axillary lymphadenopathy are also morphological predictors. The CT values of malignant lesions were higher than those of benign lesions. The cut-off value ranged from 60 Hounsfield units (HU) at 30 s [6, 7] to 90 HU on the 1-min images [8]. Optimum timing of the early phase scan was 80 s after injection of contrast media [9].

Multidetector-row computed tomography (MD-CT) detected contralateral breast cancer in 2.6% of newly diagnosed breast cancer cases [10].

Preoperative MD-CT evaluation of the extent of cancer in the breast

Extensive intraductal spread is often accompanied by invasive ductal carcinoma and becomes a major cause of positive margins after breast-conserving surgery (BCS). It is always a challenge to accurately determine the appropriate extent of resection in order to prevent local recurrence, reduce the need for re-excision, and optimize cosmetic results. Diagnostic criteria for intraductal spread using CT (axial image) are non-mass-like enhancement which is contiguous with and enhanced to the same extent as the index tumor, and the presence of linear or segmental enhancement around the main tumor [11]. The maximum