progression or intolerable toxicity. The next dose was administered only when the absolute granulocyte count was greater than 1000/mm^3, the platelet count greater than 75,000/mm^3, serum transaminase activity of no more than 100 level IU/l, and serum creatinine level of no more than 2.0 mg/dl, and when no grade 2 or higher nonhematologic toxicities except alopecia were observed. The protocol treatment was discontinued if 2 wk elapsed without fulfilling these criteria. Patients were assessed for a response after every six doses during the treatment period and every 2 mo after the completion of 18 doses.

2.3. *Response and toxicity assessment*

Response was assessed according to unidimensional measurements (Response Evaluation Criteria in Solid Tumors criteria), and toxicity was assessed according to the National Cancer Institute Common Toxicity Criteria (NCI CTC), version 2.0. Progression-free survival (PFS) was defined as time from start of therapy to disease progression, death or the most recent follow-up date; overall survival was defined as time from start of the therapy to death or the most recent follow-up date.

2.4. *Statistical analysis*

The primary end point of the trial was the partial plus complete response rate associated with weekly paclitaxel plus carboplatin in patients with bidimensionally measurable metastatic urothelial cancer. The Simon minimax design was used to plan this study on the assumption that the regimen would not be of interest if the true response rate was less than 10%, but that it would be of interest if the response rate was 30% or more. The study had a power of 80% to detect a 30% response rate. Planned accrual was the accrual of 25 eligible patients or expiration of a 2-yr period. Survival curves were estimated by the method of Kaplan and Meier, and univariate time-to-event comparisons were performed with the log-rank test. Responses according to subgroups were compared with the use of the Fisher exact test.

3. Results

3.1. *Patient characteristics*

Between May 2003 and May 2005, 35 patients with advanced transitional cell cancer were entered into this phase 2 study. Because a response was obtained in 32% of the first 25 patients, patient accrual was continued until the end of the planned 2 yr. One patient was ineligible because the patient had not received MVAC as a prior treatment. Three patients were excluded from the final analysis because they received gemcitabine monotherapy before the experimental therapy. Ultimately, 31 patients, 22 men and 9 women, were evaluable for response, toxicity, and survival (Table 1). Their median age was 67 yr (range: 51–80). Twenty-seven patients (87%) had a PS score of 0 or 1, three patients had a PS score of 2, and one patient had a PS score of 3. The site of the primary lesion was the bladder in 45% of the patients. Seventy-one percent of the patients had visceral metastasis. Nine patients (29%) had received prior MVAC as adjuvant or neoadjuvant therapy, and the other 22 patients (71%) had received it for metastatic disease. Platinum-free interval (PFI) was defined as the interval between the final dose of the prior MVAC therapy and the start of weekly paclitaxel plus carboplatin therapy. The median PFI was 4.4 mo (range: 2.5–106). In 18 patients (58%) PFI was less than 6 mo; in the other 13 patients (42%) it was 6 mo or longer. Seven patients had a PFI of more than 1 yr; only one patient had a PFI of more than 2 yr.

Table 1 – Patient characteristics (*N* = 31)

	No. of patients	%
Age, yr		
Median	67	
Range	51–80	
<70	17	55
≥70	14	45
Sex		
Male	22	71
Female	9	29
ECOG-PS		
0 or 1	27	87
2 or 3	4	13
Primary tumor site		
Bladder	14	45
Renal pelvis	9	29
Ureter	7	23
Urethra	1	3
Extent of disease		
Nodal disease only	9	29
Visceral metastasis	22	71
Lung	17	55
Liver	12	39
Bone	5	16
Prior chemotherapy		
MVAC	31	100
Adjuvant therapy	9	29
Against metastatic disease	22	71
Platinum-free interval (PFI)		
<6 mo	18	58
≥6 mo	13	42

ECOG-PS = Eastern Cooperative Oncology Group-Performance Status; MVAC = methotrexate, vinblastine, doxorubicin, and cisplatin.

3.2. *Toxicity*

The median number of doses delivered was 10 (range: 2–18). Hematologic toxicities consisted of ≥ grade 3 granulocytopenia in 18 patients (58%) (grade 3: 39%; grade 4: 19%) and ≥ grade 3 anemia

Table 2 – Toxicity analysis of evaluable 31 patients (National Cancer Institute Common Toxicity Criteria [NCI CTC], version 2.0)

Toxicity	Grade				
	0	1	2	3	4
	n (%)				
Neutropenia	7 (23)	2 (6)	4 (13)	12 (39)	6 (19)
Anemia	0 (0)	9 (29)	11 (35)	6 (19)	5 (16)
Thrombocytopenia	17 (55)	9 (29)	5 (16)	—	—
Febrile neutropenia	28 (90)	—	—	2 (6)	1 (3)
Nausea/vomiting	17 (55)	11 (35)	2 (6)	1 (3)	—
Neuropathy	9 (29)	19 (61)	3 (10)	—	—
Alopecia	7 (23)	7 (23)	17 (55)	—	—
Fatigue	17 (55)	10 (32)	4 (13)	—	—
Diarrhea	26 (84)	4 (13)	1 (3)	—	—

in 11 patients (35%); and no patients developed ≥ grade 3 thrombocytopenia (Table 2). Three patients (10%) experienced ≥ grade 3 febrile neutropenia, and the third patient enrolled whose PS score was 3 died of neutropenic sepsis within 1 mo of the final dose of chemotherapy. Subsequently we did not accrue patients with a PS score of 3.

The most common nonhematologic toxicities were alopecia (grade 1: 23%; grade 2: 55%), neurotoxicity (grade 1: 61%; grade 2: 10%), nausea and vomiting (grade 1: 35%; grade 2: 6%; grade 3: 3%), and diarrhea (grade 1: 13%; grade 2: 3%).

3.3. *Response*

Two of the 31 patients had a complete response, and 8 had a partial response. The overall response rate was 32.3% (95% confidence interval [95%CI], 15.8–48.7%) (Table 3). Among the patients whose PFI was less than 6 mo, 28% (5 of 18) had an objective response, and 38% (5 of 13) of the patients with a PFI of at least 6 mo had an objective response. The difference in the responses between subgroups according to PFI was statistically insignificant. Among the 9 patients who received prior MVAC as adjuvant or neoadjuvant therapy, 2 patients (22%) had an objective response. Among the 22 patients who received prior MVAC for metastatic disease, 8 patients (36%) had an objective response. The difference in the responses between subgroups according to the setting of the MVAC was statistically insignificant. Among the 22 patients who received prior MVAC for metastatic diseases, response rates with regard to response to prior MVAC were also analyzed (Table 4). Although responses were predominantly seen in patients who had responded to prior MVAC, one patient with resistance to prior MVAC responded to weekly paclitaxel plus carboplatin.

3.4. *Survival*

Median follow-up time was 7.8 mo. The median PFS and median survival rates were 3.7 and 7.9 mo, respectively (Fig. 1). Among the patients whose PFI was less than 6 mo, the median PFS and median survival times were 3.7 and 7.8 mo, respectively; neither survival time significantly differed from the survival times of those with PFI of at least 6 mo (median PFS: 3.3 mo; median survival: 12.4 mo). Among the patients who received prior MVAC therapy for metastatic disease, the median PFS and median survival times were 4.3 and 7.9 mo, respectively; neither survival time significantly differed from the survival times of those who received prior MVAC as adjuvant setting (median PFS: 1.6 mo; median survival: 12.4 mo).

Table 3 – Response analysis of evaluable 31 patients

	No. of patients	Response rate	
Overall response	10	32.3% (95%CI, 15.8%–48.7%)	
Complete response	2	6%	
Partial response	8	26%	
Response in PFI < 6 mo	(5/18)	28%	NS
Response in PFI ≥ 6 mo	(5/13)	38%	
Response in prior MVAC as adjuvant therapy	(2/9)	22%	NS
Response in prior MVAC against metastatic disease	(8/22)	36%	
Stable disease	12	39%	
Progressive disease	7	23%	
Not evaluable	4	13%	

95%CI = 95% confidence interval; PFI = platinum-free interval; NS = not significant; MVAC = methotrexate, vinblastine, doxorubicin, and cisplatin.

Table 4 – Response rates according to the response to prior MVAC against metastatic diseases

		No. of patients	Response n (%)
Response to prior MVAC	PR	11	5 (45%)
	SD	3	1 (33%)
	PD	5	0 (0%)
	NE	3	2 (67%)
	Total	22	8 (36%)

MVAC = methotrexate, vinblastine, doxorubicin, and cisplatin; PR = partial remission; SD = stable disease; PD = progressive disease; NE = not evaluable.

4. Discussion

Patients who had received MVAC therapy as prior treatment only in adjuvant or neoadjuvant settings and patients whose disease had progressed after MVAC therapy for metastatic disease were eligible for this phase 2 study. According to Kattan et al's report [16], when a salvage regimen included platinum, time to progression after prior platinum-based therapy, or the PFI, appeared to be important as a basis for interpreting the therapeutic efficacy of salvage treatment as well as whether the prior platinum-based therapy was for metastasis or adjuvant therapy. In this study, we defined PFI as the interval between the final MVAC therapy and the start of weekly paclitaxel plus carboplatin therapy.

Among newer active agents for urothelial cancer, gemcitabine had a 22.5% of response rate as a second-line treatment [17]. The median PFS and median survival times were 3.8 and 5.0 mo, respectively (Table 4). However, since gemcitabine has already become integrated into first-line chemotherapy [1,2], an effective second-line treatment that dose not contain gemcitabine is needed.

Paclitaxel alone yielded a 42% response rate against urothelial cancer in a first-line setting [3] but only a 10% response rate in previously treated patients [4]. Adding ifosfamide to paclitaxel had little effect, and the response rate among 13 patients who had received prior chemotherapy was only 15% [18]. Other promising new active agents are pemetrexed [19] and vinflunine [20] (Table 4), and a randomized phase 3 trial comparing vinflunine with best supportive care after progression following platinum-based chemotherapy is currently under way in Europe.

We found that the weekly paclitaxel plus carboplatin regimen in this study yielded a 32.3% response rate (95%CI, 15.8–48.7%); thus, this second-line treatment appeared to be effective against platinum-pretreated advanced urothelial cancer. This regimen was effective not only in patients with a PFI longer than 6 mo but in patients with a PFI of less than 6 mo, which indicates platinum-resistant disease. Even 28% (5 of 18) of these platinum-resistant patients had an objective response, and their median PFS and median survival times were 3.7 and 7.8 mo, respectively. In addition, 36% (8 of 22) of the patients who received prior MVAC therapy for metastatic disease had an objective response, and their median PFS and median survival times were 4.3 and 7.9 mo, respectively. Responders to weekly paclitaxel plus carboplatin include one patient who did not respond to prior MVAC therapy. These results in patients with platinum-resistant disease appear to be better than the results for weekly paclitaxel described above, which yielded a 10% response rate, and median PFS and median survival times of 2.2 and 7.2 mo, respectively [4]. We think that weekly paclitaxel and carboplatin may exert synergistic activity against advanced urothelial cancer that has

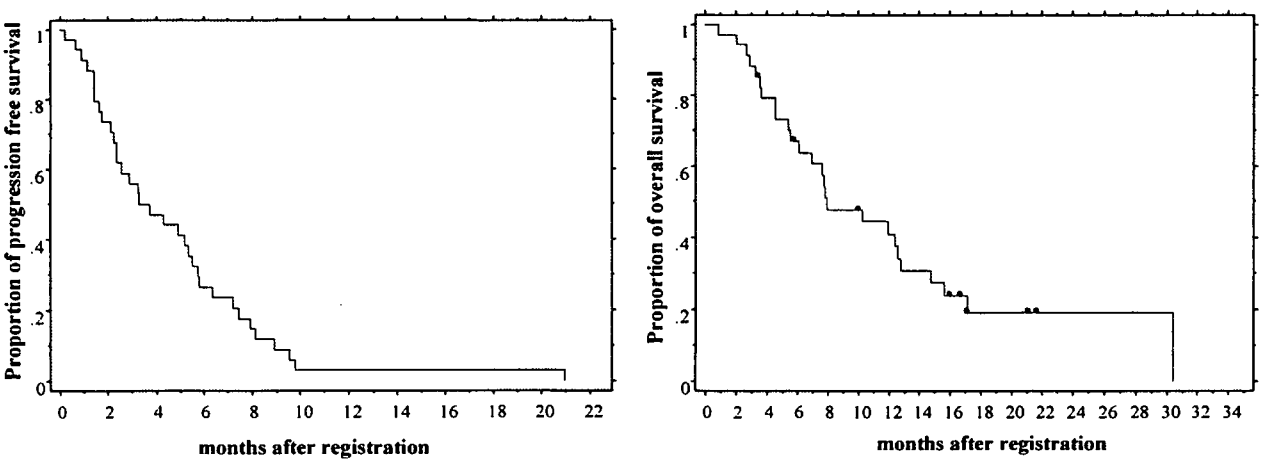

Fig. 1 – Kaplan-Meier curve of progression-free survival and overall survival.

Table 5 – Comparison of recent trials of second-line treatment of advanced urothelial carcinoma except gemcitabine combination regimen

Author, year [reference]	Treatment	No. of patients	Response rate	Median progression-free survival (mo)	Median survival (mo)
Single agent					
Witte, 1997 [22]	Ifosfamide	56	20%	2.2	5.1
McCaffrey, 1997 [23]	Docetaxel	30	13%	NR	9.0
Lorusso, 1998 [17]	Gemcitabine	31	23%	3.8	5.0
Vaughn, 2002 [4]	Paclitaxel (weekly)	31	10%	2.2	7.2
Sweeney, 2006 [19]	Pemetrexed	47	28%	2.9	9.6
Culine, 2006 [20]	Vinflunine	53	18%	3.0	6.6
Combination					
Logothetis, 1991 [24]	Fluorouracil and interferon	30	30%	NR	NR
Tu, 1995 [5]	Paclitaxel, methotrexate, and cisplatin	25	40%	NR	NR
Kattan, 1995 [25]	Ifosfamide, fluorouracil, and folinic acid	15	0%	NR	NR
Otto, 1997 [26]	Paclitaxel, carboplatin, and pertussis vaccine	18	22%	NR	NR
Sweeney, 1999 [18]	Paclitaxel and ifosfamide	26	15%	NR	8.0
De Mulder, 2000 [27]	Fluorouracil, cisplatin, and interferon	43	13%	2.3	4.9
Krege, 2001 [28]	Docetaxel and ifosfamide	20	25%	NR	NR
Di Lorenzo, 2004 [29]	Fluorouracil, folinic acid, and oxaliplatin	16	19%	NR	4.0
Vaishampayan, 2005 [21]	Paclitaxel and carboplatin	44	16%	4.0	6.0
Shinohara, 2006 [30]	Paclitaxel, ifosfamide, and nedaplatin	32	75%	8.0	22
Current series	Paclitaxel and carboplatin (weekly)	31	32%	3.7	7.9

NR = not reported.

failed platinum-containing regimens. Our results are comparable to those obtained with triweekly paclitaxel plus carboplatin in patients previously treated with platinum, which provided a 16% response rate, and median PFS and median survival times of 4 and 6 mo, respectively [21]. Furthermore, our results appear not to be inferior to the results of other second-line treatments that did not contain gemcitabine as a component of combination therapy (Table 5) [22–29]. Recently, Shinohara et al [30] reported a distinguished result for the paclitaxel, ifosfamide, and nedaplatin combination as a second-line treatment, which provided a 75% response rate, and median PFS and median overall survival times of 8 and 22 mo, respectively [30]. These data strengthen our rationale of a combination including paclitaxel and a platimum compound after failure of platinum-based chemotherapy.

Of the 31 patients, 19% experienced grade 4 granulocytopenia, 10% experienced febrile neutropenia, and 1 patient with a poor PS score died of neutropenic sepsis. With the exception of the neutropenic sepsis in the one case of toxic death, the toxicities of weekly paclitaxel plus carboplatin were all manageable. No patient experienced grade $\geq$3 thrombocytopenia, probably because of the platelet-sparing effect of paclitaxel and carboplatin [31]. In our study, no patient experienced $\geq$ grade 3 neurotoxicity, and only 10% experienced grade 2 neurotoxicity. Johannsen et al [32] recently reported $\geq$ grade 3 neurotoxicity in 6% of patients who received first-line weekly paclitaxel (100 mg/m^2) plus carboplatin (AUC 2) for advanced transitional cell carcinoma. In their study, the median number of 12 doses was administered compared with the median number of 10 doses in our study. The less frequent neurotoxicity in our study may be due to the relatively low dose of paclitaxel and the relatively low number of administrations each patient received.

5. Conclusions

Weekly paclitaxel plus carboplatin was a manageable and active second-line treatment for advanced transitional cell cancer after failure of a platinum-based regimen. Paclitaxel plus carboplatin was also effective against platinum-resistant disease, and paclitaxel and carboplatin may act synergistically.

Conflicts of interest

The authors have nothing to disclose.

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Editorial Comment on: Weekly Paclitaxel and Carboplatin against Advanced Transitional Cell Cancer after Failure of a Platinum-Based Regimen

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For many years, the standard first-line chemotherapy in metastatic transitional carcinoma of the urothelium has been the four-drug combination of methotrexate, vinblastine, doxorubicin, and cisplatin (MVAC), now replaced in most centers with gemcitabine and cisplatin (GC) with a similar efficacy but with less toxicity [1]. However, no standard has yet been established for second-line treatment. Presently, most interesting single drugs for second-line chemotherapy are gemcitabine, pemetrexed, and vinflunine. In a pooled analysis of seven studies with gemcitabine alone, an overall response rate of 25% and a complete response rate of 9% were achieved [2]. Because the efficacy seems to be independent of whether patients have received prior cisplatin-containing chemotherapy or not, gemcitabine is of potential use as second-line treatment after cisplatin-based chemotherapy not including the drug itself. In the phase 2 study of pemetrexed as second-line chemotherapy by Sweeney et al, an overall response rate of 28% was achieved [3]. This study was, however, not a clean second-line study for metastatic disease because patients with a relapse within 12 mo of adjuvant chemotherapy were also included. Presently, we are awaiting results from the randomized phase 3 study of vinflunine versus best supportive care encompassing a total of 370 patients.

In the phase 2 study by Kouno et al [4], second-line weekly paclitaxel and carboplatin resulted in an overall response rate of 32%. Nine of 31 evaluable patients were included after MVAC as adjuvant treatment. However, the response rate in the remaining patients receiving second-line treatment for metastatic disease was similar to the overall response rate. These results are interesting because paclitaxel is generally considered to be ineffective as second-line treatment following cisplatin-containing chemotherapy. Thus, this combination and schedule of paclitaxel and carboplatin deserves further evaluation.

In conclusion, well-designed studies of second-line chemotherapy for locally advanced or metastatic transitional carcinoma of the urothelium should be given high priority. In that respect, it should, however, be emphasized that patients with a primary good response to combination chemotherapy, such as MVAC or GC, and a long recurrence-free interval generally should be offered reinduction combination chemotherapy and not included in trials with new second-line drugs.

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ORIGINAL ARTICLE

Favorable outcome in patients with breast cancer in the presence of pathological response after neoadjuvant endocrine therapy ☆

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KEYWORDS
Breast cancer;
Ki-67;
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Prognostic factor

Summary Neoadjuvant endocrine therapy (NAET) can expand the number of breast cancer patients who can be treated with breast-conserving surgery and can predict benefit from adjuvant endocrine therapy. Because no validated surrogate markers for long-term outcome have been established, we conducted prospective trials to evaluate pathological response and Ki-67 index following treatment with tamoxifen or anastrozole. The study population included postmenopausal women with operable breast tumors that were both estrogen and progesterone receptor-positive and larger than 3 cm. Response was classified as pathological response (minimal response or better) and non-response. Non-responding (25.5%, vs. response 85.9%, $p = 0.002$), axillary node-positive (58.4% vs. node negative 100%, $p = 0.045$), and high pretreatment Ki-67 index (41.4% vs. low Ki-67 87.1%, $p = 0.03$) patients were significantly associated with poor 5-year relapse-free survival. Multivariate analysis of relapse-free survival indicated that pathological response was independent. Therefore, pathological response may be a favorable prognostic factor after NAET.

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Introduction

With the recent development of aromatase inhibitors, neoadjuvant endocrine therapy (NAET) has attracted attention as a potentially effective therapy that might allow breast conservation even in women with large breast tumors[1–4]. In addition, NAET offers the possibility of testing therapeutic efficacy *in vivo*, which is of great importance for optimal adjuvant treatment. However, the short history of NAET leaves several questions to be answered. First, short-term surrogate markers of subsequent risk of relapse and death from breast cancer have not been established for NAET[5]. Recently, early changes in Ki-67 have been reported to be possible predictors of long-term outcome[6–8]. The short-term reduction in Ki-67 levels in NAET (in the IMPACT trial) paralleled that observed in patients who received the same endocrine therapy in the adjuvant setting (ATAC); this suggested that the changes in Ki-67 in NAET might be predictive of long-term outcome[7]. However, these data were not obtained in direct long-term follow-up studies of NAET. Second, classifications of pathological therapeutic response, which have been mainly produced based on pathological changes following chemotherapy or radiotherapy, have not been validated for tumors treated by NAET. We conducted a small study to clarify the significance of the classification of pathological therapeutic response and the Ki-67 index as prognostic factors of long-term outcome in response to NAET.

Patients and methods

This analysis includes 45 postmenopausal women with operable estrogen and progesterone receptor (ER and PgR)-positive breast tumors that were larger than 3 cm as confirmed by core needle biopsy. These women were enrolled in two-phase II studies on NAET at the National Cancer Center Hospital (NCCH), Tokyo. Between February 1999 and July 2002, 31 patients were enrolled in a neoadjuvant tamoxifen study (neo TAM), in which they received tamoxifen for 4 months preoperatively. Between November 2002 and 2004, 17 patients were enrolled in a neoadjuvant anastrozole study (neo ANZ), in which they received anastrozole for 5 months preoperatively. Three patients in the neo TAM group were excluded from this analysis because they received preoperative chemotherapy following NAET and their tumors could not be evaluated for pathological response to endocrine therapy; two of these patients rejected mastectomy when there was no reduction of their tumors by NAET. These patients received chemotherapy with the hope that their tumors might shrink enough to allow breast-conserving surgery. Unfortunately, their tumors remained widespread in a mosaic pattern and they finally agreed to mastectomies. The third patient showed progressive disease, which led to skin invasion, and received chemotherapy before surgery. All patients provided written informed consent for study participation as approved by the institutional review board of the NCCH. Patients who responded to NAET continued the same endocrine therapy postoperatively for 5 years. Patients who showed clinically progressive disease or stable disease and pathological lymph node involvement after NAET received adjuvant chemotherapy, if tolerable, with a regimen containing anthracycline or classical CMF (cyclophosphamide, methotrexate, and 5-fluorouracil) following surgery. All patients who underwent breast-conserving surgery received postoperative radiotherapy to the ipsilateral breast.

Tumor response

Primary tumors were clinically assessed every month. Clinical complete response (cCR) was defined as the clinical disappearance of the tumor at the end of NAET, and clinical partial response (cPR) was defined as a $\geqslant$70% decrease from baseline of the largest diameter[9]. Clinical progressive disease was defined as a $\geqslant$20% increase from the most reduced size of the largest diameter. If progressive disease was observed, patients immediately underwent radical mastectomy.

Outcome measures

Relapse-free survival (RFS) was defined as the time from the initiation of treatment to local, regional, or distant treatment failure.

Histological examination

Evaluation of ER and PgR status was by immunohistochemical studies using antibodies 1D5 and PgR636 (DAKO, Glostrup, Denmark), and tumors with more than 10% strongly stained nuclei were described as ER- or PgR-positive. Tumors obtained by core needle biopsy judged as positive for both receptors before treatment were eligible for this study. HER2 status was evaluated immunohistochemically using HercepTest (Dako), and 3+: strong complete membrane staining in $>$10% of tumor cells was defined as positive.

Ki-67 was stained using the MIB-1 antibody (DAKO) according to previously described methodology[10]. Ki-67 was scored as the percentage of positively stained cells among 1000 malignant cells in specimens obtained by either core needle biopsy before treatment (baseline) or by surgery after NAET. The cut-off value for Ki-67 positivity was defined as the median value of the Ki-67 index in this study population. The proportional change in Ki-67 expression from baseline was calculated as (residual Ki-67 index—pretreatment Ki-67 index) × 1/pretreatment Ki-67 index[7].

Histopathological therapeutic response was classified according to the General Rules for the Clinical and Pathological Recording of Breast Cancer 2005[11]. For Grade 0, no response was observed; Grade 1a comprised those tumors with mild changes in cancer cells regardless of the area, or marked changes seen in less than one-third of cancer cells; Grade 1b comprised tumors with marked changes seen in more than one-third but less than two-thirds of tumor cells; Grade 2 tumors contained marked changes in more than two-thirds of tumor cells; and Grade 3 tumors demonstrated a complete response, with no cancerous cells remaining. Mild changes include slight degenerative changes in cancer cells not suggestive of cancer cell death (including cancer cells with vacuolation of the cytoplasm, eosinophilic cytoplasm, swelling of the nucleus, etc). Marked changes include marked degenerative changes in cancer cells suggestive of cancer cell death (including liquefaction, necrosis, and disappearance of cancer cells). The pathological response group was defined as tumors with Grade1a, 1b, and 2 responses. The non-response group was defined as tumors with Grade 0 response.

Statistical analysis

The χ^2 test was used for comparisons of tumor characteristics and responses among groups. The Kaplan–Meier methods were used to generate RFS curves. The log rank test was used for the comparison of RFS between two groups. Differences with $p<0.05$ were considered to be significant.

Table 1 Characteristics of patients and tumors treated with tamoxifen (neo TAM group) and anastrozole (neo ANZ group).

	Neo TAM group ($n=28$)	Neo ANZ group ($n=17$)	
Age	60 (51–75)	61 (54–87)	
Tumor before NAET			
T2	18	11	
T3	7	4	NS
T4	3	2	
Clinical response			
CR	1	3	
PR	12	10	
NC	15	4	$p=0.05$
PD	0	0	
Surgery			
Mastectomy	17	13	
BCS	11	4	NS
Pathological response			
Grade 2	3	3	
Grade 1b	4	2	
Grade 1a	11	11	$p=0.02$
Grade 0	10	1	
Axillary nodal status			
Negative	7	6	
1–3	12	7	NS
4–9	7	3	
>10	2	1	

NAET: neoadjuvant endocrine treatment; CR: complete response; PR: partial response; NC: no change; PD: progressive disease; NS: not significant; BCS: breast-conserving surgery.

Results

Tumor and patient characteristics in the neo TAM and neo ANZ groups are shown in Table 1. The clinical response rates (cCR+cPR) for the neo TAM and neo ANZ groups were 46.4 and 76.5%, respectively. Of the neo ANZ group, only four patients underwent breast-conserving surgery, because some patients with good clinical responses chose mastectomies and refused postoperative radiotherapy. Patients treated with neo ANZ showed a statistically significantly higher rate of pathological

Table 2 Tumor characteristics and responses to NAET stratified by patients with events and those without events.

	Non-response group ($n = 11$)	Pathological response group ($n = 34$)	
Age	57 (51–73)	61 (52–87)	
Tumor before NAET			
T2	9	20	
T3	1	10	
T4	1	4	NS
Histological grade before NAET			
Grade 1	1	8	
Grade 2	6	15	
Grade 3	4	9	NS
Not available	0	2	
HER2 status before NAET			
Negative	11	34	
Positive	0	1	NS
NAET			
Tamoxifen	10	18	
Anastrozole	1	16	NS
Clinical response			
CR	0	4	
PR	4	18	
NC	7	12	NS
PD	0	0	
Ki-67 index before NAET			
High	6	17	
Low	5	17	NS
Residual Ki-67 index			
High	7	16	
Low	4	18	NS
Proportional reduction of Ki-67 index			
Median(Q_1–Q_3)	−0.05 (−0.67–0.37)	−0.46 (−0.85–0.83)	NS
Lymphovascular invasion			
Negative	9	28	
Positive	2	6	NS
Axillary nodal status			
Negative	2	11	
1–3	6	13	
4–9	1	9	
>10	2	1	NS
Adjuvant therapy			
Endocrine only	5	20	
Chemotherapy added	6	14	NS

Q_1: first quartile; Q_3: third quartile.

response (Grades 1+2) than those treated with neo TAM ($p = 0.02$).

Tumor characteristics stratified by patients with pathological response or non-response are shown in Table 2. There were no statistically significant differences in tumor size, histological grade, HER2 status, clinical response, lymphovascular invasion, pathological nodal status, or addition of adjuvant chemotherapy between these groups. Reduction of Ki-67 was not significantly associated with either pathological or clinical response.

The median follow-up time after NAET was 44.7 months. There were 11 locoregional and/or metastatic events during this time. No ipsilateral breast tumor recurrence was observed after breast-conserving surgery. Patients with pathological non-response (25.5%, vs. response group 85.9%, $p = 0.002$; Fig. 1), axillary node positivity (58.4% vs. node negative 100%, $p = 0.045$), addition of adjuvant chemotherapy (41.2% vs. only endocrine therapy 77.5%, $p = 0.01$), and high pretreatment Ki-67 index (41.4% vs. low Ki-67 index 87.1%, $p = 0.03$; Fig. 2) were significantly associated with poor 5-year RFS. Initial T category, histological grade, clinical response, type of endocrine therapy, presence of reduction in Ki-67 values, and lymphovascular invasion was not associated with survival.

The median follow-up time for the neo TAM group was 65.8 months. In this group, patients with pathological non-response (28.0%, vs. response group 88.2%, $p = 0.006$; Fig. 3), axillary node positivity (59.9% vs. node-negative 100%), addition of adjuvant chemotherapy (43.2%, vs. only endocrine therapy 77.8%, $p = 0.03$), and high residual Ki-67 index (44.0%, vs. low Ki-67 index 100%, $p = 0.01$) were significantly associated with poor 5-year RFS.

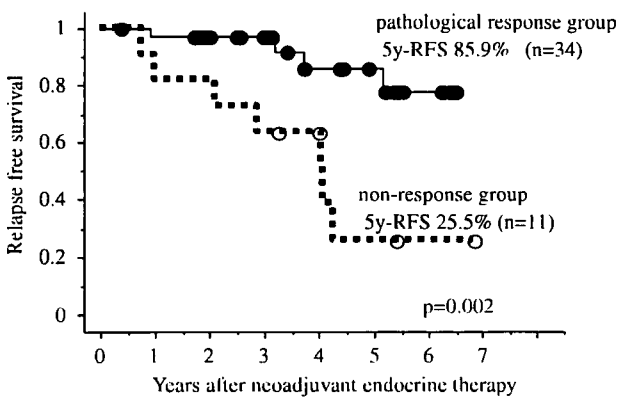

Figure 1 Relapse-free survival curves following neoadjuvant endocrine therapy stratified into a pathological response group (—) and a non-response group (- - -). A statistically significant difference was observed between the groups ($p = 0.002$).

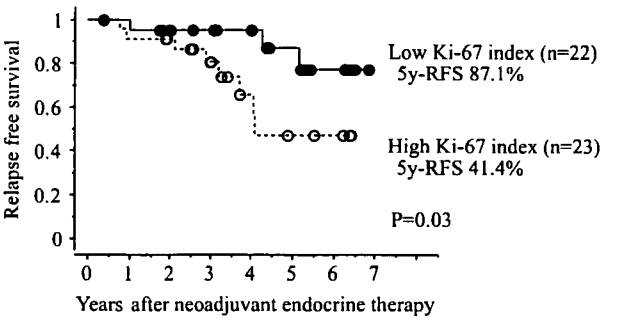

Figure 2 Relapse-free survival curves following neoadjuvant endocrine therapy stratified into a low pretreatment Ki-67 index group (—) and a high Ki-67 index group (- - -). A statistically significant difference was observed between the groups ($p = 0.03$).

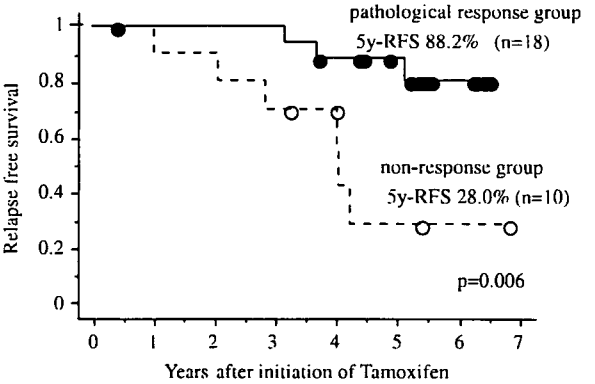

Figure 3 Relapse-free survival curves following neoadjuvant endocrine therapy using tamoxifen stratified into a pathological response group (—) and a non-response group (- - -). A statistically significant difference was observed between the groups ($p = 0.006$).

The median follow-up time for the neo ANZ group was 30.0 months. The pathological response group achieved statistically better 3-year RFS than the non-response group (93.3% vs. 0%, $p<0.0001$).

Multivariate regression analyses using a logistic regression model were conducted to identify independent prognostic factors for RFS (Table 3). These analyses indicated that pathological response ($p = 0.007$) was significantly related to RFS.

Discussion

Although the sample sizes in this study are small, the pathological response group showed significantly more favorable outcomes than the non-pathological response group following NAET. This result is supported by all of the analyses conducted in this study and suggests that the pathological therapeutic response may be a prognostic factor for

Table 3 Multivariate analysis for RFS after NAET.

		Hazard ratio (95%CI)	*p*-value
Pathological response	Non-response/response	6.3 (1.6–23.8)	0.0067
Pretreatment Ki-67	Low/high	0.26 (0.055–1.17)	0.079
Residual Ki-67	Low/high	0.65 (0.14–2.98)	0.58

RFS: relapse-free survival; CI: confidence interval.

long-term outcome following NAET. The response necessary for a favorable prognosis seems to differ between neoadjuvant chemotherapy and NAET. In the neoadjuvant cytotoxic chemotherapy setting, where response (pCR or not) is a clinically significant predictor of outcome[12], long-term outcome following treatment with cytostatic agents can be predicted based on the achievement of minimal pathological change. Using chemotherapy, total killing of cancer cells is necessary to improve prognosis; therefore, physicians should pursue regimens that will reach the highest pCR rates possible. On the other hand, only a few patients have been reported to achieve pCR following NAET[3]. This is one reason for hesitation in using endocrine agents in a neoadjuvant setting. However, with endocrine therapy, minimal pathological changes may have the same power to improve prognosis.

In this study, low Ki-67 index before NAET in all cases and low residual Ki-67 index in the neo TAM group were significant favorable prognostic factors. Ki-67 has been reported to carry modest prognostic significance and the residual (after treatment) level of Ki-67 may be a better predictor of response and/or absolute long-term outcome than the proportional reduction in Ki-67 because it is more likely to relate to the growth rate of the persistent disease[13]. The results of this study are concordant with these results. The results of the IMPACT trial supported the hypothesis that a reduction of Ki-67 in NAET might be predictive of long-term outcome, but this was not demonstrated in this study. As Urruticoechea has reported that a change in Ki-67 score of at least 32–50% between two determinations using core needle biopsies is required to consider the difference statistically different for an individual patient and attributable to treatment effects[13], the problem with the reproducibility of Ki-67 measurements must be overcome.

Patients who underwent additional adjuvant chemotherapy showed a statistically significant reduction in RFS compared with those who underwent only endocrine therapy. Selection bias must be considered, as most of the patients with positive lymph nodes were treated with chemotherapy. However, whether or not the chemotherapy was efficacious remains controversial because hormone-sensitive breast cancer is less responsive to chemotherapy[14,15]. Further investigations are required to determine the best treatment plan for such cases.

Neoadjuvant chemotherapy has now been established as one of the standard treatments for operable breast cancer. On the other hand, there is less evidence on NAET than on neoadjuvant chemotherapy, including long-term outcome. In this situation, NAET should be used to treat selected patients who will obtain great benefit from endocrine therapy and will not respond to chemotherapy and/or do not need chemotherapy. Without a doubt, hormone receptor status is the first eligibility criterion. Many studies on neoadjuvant chemotherapy have confirmed that hormone-sensitive tumors show worse responses to chemotherapy than hormone-resistant tumors[14,15]. However, not all hormone-sensitive tumors respond to endocrine therapy, underscoring the need for additional predictive tests. Gene analysis can be used as a second eligibility criterion. A multigene assay (Oncotype DX)™ succeeded in predicting that approximately half of the women with node-negative, hormone receptor-positive breast cancer who were treated with local therapy and tamoxifen have an excellent prognosis, with more than 90% having 10-year relapse-free survival; these patients are unlikely to benefit from chemotherapy[16,17]. A more favorable response and long-term outcome without severe adverse events may be achieved with only hormone therapy using gene expression profiles to select patients who are good candidates for NAET.

This study suggests that pathological response is a favorable prognostic factor following NAET. We await validation of these results in large studies such as the IMPACT trial or Letrozole P024 to establish the surrogate markers that predict the risk of recurrence.

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Review Article

Current Trends and Controversies over Pre-operative Chemotherapy for Women with Operable Breast Cancer

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The multi-disciplinary approach, including surgery, chemotherapy, endocrine therapy and radiation therapy, has become the standard treatment for primary breast cancer patients. The indication of pre-operative chemotherapy has been extended to women with potentially operable breast cancer based on the results of large randomized studies and has become an attractive option that extends the chance of breast conservation. The clinical and pathological responses to pre-operative chemotherapy correlates with long-term outcome. The anthracycline-containing regimen is now considered the standard. Sequential administration of non-cross-resistant drugs, namely taxanes, improves local tumor response but its long-term benefit has been controversial. Prediction of response to pre-operative chemotherapy still remains a challenge. Identification of useful predictive markers and development of molecular-targeted drugs is the key to individualized therapy in the future.

Key words: pre-operative chemotherapy – breast cancer – advantage – response – long-term outcome – prediction

INTRODUCTION

The multi-disciplinary approach, including surgery, chemotherapy, endocrine therapy and radiation therapy, has become the standard treatment for primary breast cancer patients with a high risk of recurrence. Although mortality from breast cancer is decreasing in western countries thanks mainly to early detection of the disease by mammography screening and wide usage of post-operative adjuvant systemic therapy (1), its incidence and mortality are steadily increasing in the rest of the world, including Japan (2).

When it first emerged in late 1970s, the use of pre-operative (primary) chemotherapy had been primarily limited to women with inoperable locally advanced breast cancer to enable optimal local therapy (3–5). Later on, large randomized trials proved that pre-operative chemotherapy has at least the same survival benefit as the post-operative chemotherapy (6), and its indication has been extended to women with potentially operable breast cancer.

However, with long-term survivors increasing by systemic therapy in early breast cancer, the 'survivorship' or importance of quality of life after primary therapy has recently come into the limelight. Whether an attempt at breast conservation can be made at the time of definitive surgery is one of the important issues discussed among patients and physicians. Pre-operative chemotherapy is an attractive option for those who have large tumors but a strong interest in breast conserving surgery.

In this review, we describe available evidence and discuss current controversies and future prospects of pre-operative chemotherapy, taking account of its two major clinical roles; eradication of micrometastatis and increased chance of breast conservation.

RATIONALE OF PRE-OPERATIVE CHEMOTHERAPY

Biologic rationale for pre-operative adjuvant chemotherapy was derived from the pre-clinical studies in animal models. It had been known that growth kinetics of metastatic tumors change after surgical removal of the primary lesion (7). The greatest effect of chemotherapy was observed when it was administered prior to operation (8, 9). These observations led to a hypothesis that early systemic chemotherapy prior to surgery might further reduce the risk of metastasis.

The landmark trial in a clinical setting was the National Surgical Adjuvant Breast and Bowel Project (NSABP)

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B-18 trial, which showed pre-operative chemotherapy for operable breast cancer by doxorubicin 60 mg/m^2 and cyclophosphamide 600 mg/m^2 (AC) was at least as effective as post-operative adjuvant chemotherapy with the same regimen in terms of disease-free and overall survival (10). The results were consistent over a longer follow-up period (6) and the result of another large randomized trial conducted in Europe was also confirmatory (11). A recent meta-analysis of pre-operative and post-operative chemotherapy (partly including T4 disease) indicated that pre-operative chemotherapy was equivalent to post-operative therapy in terms of survival and disease progression (12).

Thus the available clinical data has not demonstrated a convincing difference in long-term outcome as hypothesized in pre-clinical studies. However, a higher proportion of women were able to undergo breast conservation surgery. In addition, because the extent of clinical and pathological responses to pre-operative chemotherapy correlates with survival (10), improved tumor response in this setting is expected to improve the overall outcome.

ADVANTAGE OF PRE-OPERATIVE CHEMOTHERAPY

The advantage of pre-operative therapy is that one can subjectively evaluate the response to systemic therapy *in vivo*. Both clinical and pathological responses have been associated with prolonged disease-free and overall survival (6, 8) and they are used as the primary endpoint in clinical trials. Unlike post-operative adjuvant chemotherapy, one can avoid or minimize the unnecessary toxicities from cytotoxic agents by changing treatment strategy when the tumor is not responding to a certain regimen.

Pre-operative chemotherapy is an attractive option for women who wish to reduce the extent of local surgery. Clinical trials provide evidences that 28–89% of women can undergo breast conserving surgery when they might not be otherwise qualified (12).

Because breasts are located on the body surface, one can easily obtain the tumor cells or tissue by either fine needle aspiration or core needle biopsy with minimal invasions. As one can also evaluate the response to systemic therapy in a subjective manner and because patients are usually chemotherapy naïve, a pre-operative setting can be an ideal *in vivo* laboratory for biomarker studies using tumor specimens.

DISADVANTAGE OF PRE-OPERATIVE CHEMOTHERAPY

The overall response rate of pre-operative chemotherapy is 75% on average (range 49–100%), whereas fewer than 5% of the patients with operable breast cancer progress during pre-operative chemotherapy and some more do not even show major responses (13). For such patients with progression, the delay of local treatment may be of disadvantage at least in terms of local control. Pre-operative chemotherapy is also associated with significantly increased risk of loco-regional disease recurrence (12).

Another potential disadvantage of pre-operative chemotherapy is the loss of initial histological information such as tumor size, nodal status and biologic markers. According to the current guidelines, application of post-operative chemotherapy is to be decided by weighing the baseline risk, endocrine responsiveness and estimated risk reduction and harm of the treatment (14). Risk of recurrence is estimated based on the clinical and pathological information obtained from surgical specimens. In a pre-operative setting the information on tumor size and nodal status will inevitably be imprecise and intra-tumor heterogeneity of histologic type, histologic grade and biomarker expression cannot be taken into account. It may potentially put patients into danger of over- or under-treatment. Currently, core-needle biopsy is mandatory prior to pre-operative chemotherapy to obtain as much pre-treatment histopathological information as possible.

TREATMENT REGIMENS

Using clinical or pathological responses as surrogate endpoints of overall survival, optimal systemic therapies have been investigated in pre-operative settings in patients with early breast cancer. The general consensus reached is that an anthracycline-containing doublet (doxorubicin or epirubicin with cyclophosphamide) or triplet (doxorubicin or epirubicin with cyclophosphamide and 5-fluorouracil) should be used as the initial chemotherapy strategy for pre-operative chemotherapy (15, 16).

The sequential use of non-cross-resistant agents is likely to augment the response of pre-operative chemotherapy (17, 18), among which taxanes are the most investigated drug. Overall, results of randomized trials indicate that the incorporation of taxane increases the rate of pathological complete response (pCR) by 6–16% compared to anthracycline/cyclophosphamide-based regimens (19, 20). Smith et al. randomized patients who achieved clinical response to the initial four cycles of cyclophosphamide/vincristin/doxorubicin/predonisone (CVAP) therapy to receive further four cycles of CVAP or four cycles of docetaxel (Aberdeen trial) (21). The sequential use of docetaxel resulted in enhanced clinical and pathological responses even in anthracycline-sensitive tumors. In NSABP-B27 trial, the addition of four cycles of docetaxel after pre-operative AC increased the clinical complete response rate (40% versus 63%), clinical overall response rate (86% versus 91%) and the pCR rate (14% versus 26%) compared with pre-operative AC therapy alone (20). However, the addition of taxane in pre-operative or post-operative setting after AC did not improve the long-term outcome in this trial (22).

Treatments incorporating molecular-targeting drugs are of interest. Trastuzumab is effective for patients with advanced

breast cancer over expressing HER2 (23). In adjuvant settings, at least one year of trastuzumab given sequentially or concomitantly with chemotherapy significantly improves disease-free and overall survival (24, 25). Moreover a short course (9 weeks) of trastuzumab administered concomitantly with docetaxel or vinorelbine seems to be effective in HER2-positive subset of patients in adjuvant settings (26).

For pre-operative settings, there are a limited number of phase II studies reporting the use of trastuzumab (25, 27, 28). The only randomized trial reported was by Buzdar et al., who compared neoadjuvant chemotherapy for HER2-positive, operable breast cancer with or without administration of trastuzumab (29). This study was closed by the recommendation of Data and Safety Monitoring Board of the institution according to early-stopping rule, because pCR rate, the primary endpoint, was strikingly superior in the chemotherapy plus trastuzumab arm (given simultaneously for 24 weeks) compared with the chemotherapy-alone arm (65% versus 26%, $p = 0.016$). We still need to confirm if this significant difference in pathological response will be translated into prolonged overall survival by long-term follow-up and also the cardiac safety of trastuzumab in combination with chemotherapy should be assessed.

CONTROVERSIES OVER PRE-OPERATIVE CHEMOTHERAPY

Evaluation of Residual Tumor for Optimal Surgery

Optimal imaging modality has not been established to definitely localize the remaining tumor. Usually, serial imaging studies are performed before and after pre-operative chemotherapy. Magnetic resonance imaging or computerized-tomography scanning may supplement conventional breast imaging studies by mammography and ultrasonography (30–33).

The use of functional imaging techniques such as fluorine-18 fluorodexyglucose positron emission tomography ([^{18}F]-FDG PET) is of interest for the evaluation of therapeutic response to systemic therapy in breast cancer. The change in [^{18}F]-FDG uptake reflects the alteration in cellular glycolysis. Some relatively small studies reported that [^{18}F]-FDG PET after a single pulse of chemotherapy predicted pCR or minimal residual disease with a sensitivity of 85–100% and a specificity of 74–85% (34–36). FDG-PET is promising for clinical application in future to detect non-responding tumor to avoid unnecessary toxicities from cytotoxic therapy.

Feasibility of Sentinel Lymph-Node Biopsy (SNB) in Patients Treated with Pre-operative Chemotherapy

Axillary staging by SNB may allow omission of axillary dissection in sentinel-node negative patients without compromising the long-term outcome (37). However the optimal timing and feasibility of SNB in the setting of pre-operative chemotherapy have not been established.

Identification rate of SNB following pre-operative chemotherapy are reported to be 84–93% and 78–93%, in single-institution series and multi-center studies (38), respectively. High false-negative rates up to 25–33% have been reported for several small single institution studies (39, 40), but in multi-institutional studies using radiocolloid with or without blue dye, false-negative rates range between 5 and 13% (38), which are similar to those observed when it was carried out before systemic chemotherapy.

There still remain concerns about the use of SNB following chemotherapy in patients with clinically positive axilla (41), SNB after chemotherapy possesses a potential to maximize the benefit of axillary downstaging by pre-operative systemic treatment, in other words, avoidance of complications related to axillary dissection and decision-making of adding further chemotherapy.

Alteration of Biological Markers

The changes in the expression of hormone receptors and HER2 protein during pre-operative chemotherapy may influence the clinical decision of adjuvant hormonal and trastuzumab therapy. In studies using immunohistochemistry, the administration of pre-operative chemotherapy did not alter the expression patterns of HER2 and hormone receptors (42–45).

However, a study was conducted to compare gene expression profile of pre-treatment biopsy specimens with those in tumors remaining after doxorubicin-containing pre-operative chemotherapy using DNA array. There were differences in the gene expression profile in tumors that showed a response, but not in tumors that did not respond to therapy (46). Biological and clinical implications of the change of gene expression profile in responding tumors need further elucidation.

Definition of Pathological Response

Primary systemic treatment is increasingly recognized as the best model for the quick development of new treatment strategies in early breast cancer. pCR after pre-operative chemotherapy has been chosen as the primary endpoint of clinical trials, because it is validated as the surrogate marker of improved outcome (47, 48). However, diverse definitions of pathological response are used by different investigators (10, 47, 49–53). Some of these grading systems allow inclusion of residual ductal caricinoma *in situ* (DCIS) without invasive component in the definition of pCR. However, there is no confirmatory data to justify the concept that there is no difference in prognosis between patients with no invasive or *in situ* disease and those with residual DCIS. Jones et al. investigated whether the prognosis for patients with residual DCIS is the same as that for patients with no residual tumor cells, but could not demonstrate significant

prognostic difference (54). However, this study was statistically underpowered to draw any conclusions.

Ideally, response to chemotherapy should be measured as a continuous variable. No system satisfies the need of accurate pathologic evaluation for the majority of patients who achieve partial or minor response to pre-operative chemotherapy. Rajan et al. proposed that the product of residual tumor size and cellularity might be a more clinically relevant indicator of tumor response than assessing tumor size alone (55). Though it is an interesting proposal, the method needs to be validated in correlation with long-term outcome.

Outcome after Pre-operative Chemotherapy and Surgery

Several studies have attempted to find more accurate predictors for survival after pre-operative chemotherapy than pCR in the primary tumor. This is because substantial risk of systemic recurrence still remains even if pCR is achieved, whereas substantial patients have excellent prognosis even if pCR is not achieved. If the long-term risk is high, they will be the candidates for clinical trials to determine whether additional aggressive therapy will be of benefit. If a good prognosis is expected even without good response to pre-operative therapy, aggressive chemotherapy might be over-treatment in pre-operative setting.

In the report of retrospective studies from Royal Marsden Hospital and M. D. Anderson Cancer Center, pathologically negative axillary lymph nodes after pre-operative chemotherapy, not pCR in the primary tumor, remained the independent prognostic factor for disease-free survival and overall survival in multivariate analysis adjusted for other prognostic factors (56–58).

It was revealed by a retrospective multivariate analysis of the clinicopathological factors of the 226 patients who had pCR after pre-operative chemotherapy that pre-operative clinical stage IIIB, IIIC, and inflammatory breast cancer, axillary lymph nodes more than 10, and pre-menopausal status were the independent prognostic factors of distant metastasis (59). In another study, only histological grading had an independent prognostic impact on disease-free and overall survival after adjustment for pCR to pre-operative chemotherapy containing doxorubicin (60). Carey et al. found that American Joint Committee on Cancer Tumor-Node-Metastasis staging after pre-operative chemotherapy was useful in prediction of distant disease-free survival and overall survival (61).

Rouzier et al. constructed nomograms combining clinical variables associated with pCR that might accurately predict pCR and distant disease-free survival (62). This was confirmed in an independent dataset within the study. The nomogram included size of residual tumor and the number of metastatic nodes at the time of surgery, histologic grade, estrogen receptor (ER) status and histologic type. On the other hand, biologic markers such as expression of HER2 (63), EGFR (64), p53 (65) or MDR1 gene (66) in tumor specimen before pre-operative chemotherapy, reduction of expression in topoisomerase II-α (70) or MLH1 (71) after pre-operative chemotherapy are suggested to predict long-term outcome. Although it is not known whether these markers would add to or replace the nomogram, development of more accurate and comprehensive tools for prediction of prognosis is awaited.

Prediction of Response to Pre-operative Chemotherapy

The pre-operative setting is ideal to explore molecular predictors of response to therapy. Various clinical and pathologic variables have been studied. Among them, ER status, histologic grade and smaller tumor size seem to be associated with the response to pre-operative chemotherapy (47, 69).

In previous retrospective studies, clinical and pathological responses to pre-operative chemotherapy appear to be lower in invasive lobular carcinoma (ILC) as compared to invasive ductal carcinoma (IDC), and patients with ILC were more likely to receive mastectomy after initial attempt for breast conservation (70–73). However, low pCR rates in ILC have not been translated into survival disadvantage (70–72). These data suggest that different approach should be taken in the clinical management of patients with ILC.

In a biomarker study, ER expression, absence of HER2 and a decrease in Ki67 correlated with good clinical responses subsequent to a pre-operative chemoendocrine therapy (74). Among other biomarkers, bcl-2 and p53 have been studied. bcl-2 has been shown to protect cells from apoptosis induced by chemotherapeutic drugs (75). Although high expression of bcl-2 has been hypothesized to play a role in resistance to chemotherapy, it is still controversial. In one study, higher bcl-2 expression at diagnosis was predictive of pCR in univariate analysis but it did not retain its impact in multivariate analysis (76), while other studies did not find any correlation between bcl-2 expression and the response (77, 78).

p53 is also a potential predictive marker. Active p53 promotes apoptosis in growth-arrested cells whereas loss of p53 function has been reported to enhance cellular resistance to various chemotherapeutics (79). In a clinical setting, in patients treated with single agent epirubicin, mutant p53 was a significant predictor for poor clinical response, but the association was weaker in patients treated with cyclophosphamide/methotrexate/5FU with or without tamoxifen (65). Another study demonstrated that a tumor expressing wild-type p53 was related to resistance to single agent doxorubicin therapy in multivariate analysis (80). *TP53* gene mutation and over expression of p53 were related to epirubicin-containing chemotherapy, but response to paclitaxel seemed to be related to p53-negative tumors (81).

Tumor response and toxicities are different among individual patients. Pharmacogenomic studies aim to elucidate the genetic bases for inter-individual differences and to enable individualization of care. DNA microarray is one of the modern high-throughput biotechnologies that allow

researchers to analyze expression of multiple genes in concert and relate the findings to clinical parameters. In breast cancer, several groups have reported preliminary results suggesting that the gene expression profile of the primary tumor may predict the tumor's response to pre-operative chemotherapy (82–86). One major limitation of microarray studies is overfitting of the predictior: the number of mRNA transcripts far exceed the number of samples (87, 88). The accuracy of the predictive model is low in independent data set (89). More rigorous and critical evidence is necessary before multi-gene predictors can be accepted as a useful and reliable tool in clinical practice.

Pre-operative Endocrine Therapy

The relative benefit of chemotherapy is less in endocrine-responsive disease as compared with endocrine non-responsive disease (1) and recent consensus of the clinical community lays emphasis on the endocrine responsiveness in decision-making of adjuvant systemic therapy (14). Pre-operative endocrine therapy is an attractive alternative for endocrine-responsive disease, because it is easy to perform and can also avoid acute and late side effects caused by cytotoxic chemotherapy, but pre-operative endocrine therapy has not been accepted as the standard therapy because of the slow rate of response (90). We need more accurate measures to select the patients who are most likely to respond to endocrine therapy without compromising the potential benefit of chemotherapy.

Application to Molecular-Targeted Therapy

Molecular-targeted drugs are anticipated to individualize the therapeutic strategy based on the biology of the tumor. To date, the presence of a target still does not satisfactorily guarantee a response to therapy, but efforts are being made to elucidate the key components of the molecular pathways targeted by a specific agent.

Moshin et al. reported a pre-operative study of trastuzumab as a single agent in HER2-positive locally advanced breast cancer (91). They administered trastuzumab as a single agent for the first 3 weeks, followed by a combination of trastuzumab and docetaxel. Of note, partial response was observed in eight among 35 patients after only 3 weeks of trastuzumab. The accompanying biomarker study suggested that the main mechanism of action of trastuzumab is inhibition of the PI3K/Akt pathway, which results in an increase of apoptosis (79). The clinical role of single-agent trastuzumab in HER2-positive tumors has not been determined, but it is attractive if we can select the responders to trastuzumab as this is usually less toxic than cytotoxic chemotherapy.

A report by Polychronis et al. is unique in respect of testing the efficacy of combination of targeted therapy based on biology-derived hypothesis (92). It was a double-blind placebo controlled phase II randomize trial of pre-operative gefitinib versus gefitinib versus anastrozole in post-menopausal patients with ER- and EGFR-positive primary breast cancer. The tumors of patients assigned to combination therapy had a greater reduction of Ki67 labeling index than those assigned to gefinitib alone. Although the number of patients in this study was so small that we do not yet know whether reduction in proliferation will be translated into clinical benefit, we foresee a future of individualized therapy.

FUTURE DIRECTIONS

Pre-operative chemotherapy has become the standard of care in management of primary breast cancer. However, we should be aware that a substantial portion of patients may be over-treated by pre-operative chemotherapy because of inaccurate pre-treatment staging. In NSABP-B27 study, addition of docetaxel was beneficial in terms of disease-free survival not in complete responders or non-responders but only in partial responders in a subset analysis according to clinical response after AC. Who needs additional systemic therapy? Who can avoid systemic therapy?

Development of endocrine therapy and trastuzumab has opened the door to important therapeutic advance of 'molecular-targeted therapy'. Transcriptional profiling has revealed that expression levels of these targets, i.e. ER and HER2, are the major genetic determinants of the biology of the disease (93). Thus, we can foresee the future of systemic therapy individualized with endocrine responsiveness and involvement of HER2 signaling pathway. However, to date, the predictive value of screening test for molecular targets remains unsatisfactory.

Identification of clinically useful, prognostic and predictive molecular markers is highly anticipated to optimize therapeutic regimens. The current probability-based therapeutic strategy, 'empiric treatment' so to speak, might give way to biology-based, individualized strategy, 'marker-based treatment', when additional biologic markers are identified that make 'targeted therapy' more targeted and effective. Pharmacogenomic researches that accompany pre-operative therapy might help better understand the biology of breast cancer and thus promote the development of new therapeutic strategies.

Conflict of interest statement
None declared.

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