

Chemotherapy for breast cancer (Review)

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Abstract. The use of cytotoxic chemotherapy in both advanced and early stage breast cancer has made significant progress in the last 10 years with several landmark studies identifying clear survival benefits for newer therapies. In spite of these developments the optimal approach for any specific patient can not be determined from a literature review or decision-making algorithm alone. Treatment choices are predominantly based on practice determined by individual or collective experience and the historical development of treatment within a locality. The improvement in the understanding of the molecular biological basis of breast cancer provides possible targets for novel therapies. Personalised therapies for breast cancer based on the molecular characteristics of the tumour could improve the risk:benefit ratio of current therapies. Increased improvements in the use of a panel of biomarkers will thus not only move us towards tailored therapies but will also spare a group of patients that do not benefit from adjuvant chemotherapy. At the same time a better understanding of tumour biology will also streamline the development of new regimens for those who are unlikely to benefit from existing drugs. This review will focus on the evidence for the use of chemotherapy and highlight advances in chemotherapy treatments with the addition of new and novel drugs marching into our clinics as standard treatments based on evidence from clinical trials and from a better understanding of tumour biology that has transformed the outlook in breast cancer in both the adjuvant and metastatic setting.

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1. Introduction

With an incidence of more than 1,000,000 new cases and 370,000 deaths annually worldwide breast cancer remains a major challenge today. Despite an increasing incidence of breast cancer, the disease specific mortality has been declining in the majority of developed countries (1). The use of systemic therapy in early breast cancer is undoubtedly a major reason for that. What we find striking in breast cancer is that although the risk of distant recurrence is greatest during the first decade, it may still be significant during the second decade post diagnosis. The main aim of systemic adjuvant treatment is to control any micrometastatic disease, reduce the recurrence rate, and improve the long-term overall survival.

Since most of the improvement in 15-year breast cancer mortality produced by adjuvant chemotherapy and hormonal therapy and by adjuvant radiotherapy (2) occurs after the first 5 years, there may be a delay of a decade or so between any widespread changes in practice and the main effects that these will eventually have on national breast cancer mortality rates (3). Thus, earlier diagnosis, wider use of adjuvant treatments, or both, during the 1980s contributed significantly to the sudden decreases of 25-30% noted in the USA and UK breast cancer mortality rates (4). Further moderate improvements during the 1990s involving better local disease control (partly because of more careful and more extensive screening) and better use of systemic treatments both for early and for advanced disease should in total help these decreases in national mortality rates to continue throughout the present decade (5). Despite improvements with better understanding of the use of adjuvant therapies for early stage breast cancer, the treatment of metastatic disease remains a major challenge. The use of anthracyclines and taxanes in the adjuvant setting has led to an increasing number of women presenting with metastatic disease having already been exposed to these agents adding to the complexities of their management. Despite being incurable, metastatic breast

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cancer (MBC) often remains chemosensitive such that symptom control and prolongation of survival can be achieved. However, response duration remains disappointingly short and long-term survival remains uncommon. Breast cancer remains a classic model where chemotherapy treatments have been tested in advanced metastatic settings and having shown efficacy with tolerable toxicity have marched into the adjuvant setting.

The ongoing expansion in the understanding of the molecular biological basis of breast cancer provides further potential targets for novel therapies.

The development of trastuzumab, a humanised monoclonal antibody against HER-2/neu provides the first example of a rationally designed targeted biological therapy for breast cancer successfully tested in large randomised clinical trials (6) and is now widely accepted as standard therapy. In spite of these key developments, resistance to therapy remains a challenge in the management of advanced breast cancer (3). The improvement in the understanding of the molecular biological basis of breast cancer provides possible targets for novel therapies. Personalised therapies as adjuvant (or neo-adjuvant) chemotherapy for breast cancer based on the molecular characteristics of the tumour could improve the risk-benefit ratio of current therapies. Increased improvements in the use of a panel of biomarkers will thus not only move us towards tailored therapies but will also spare a group of patients that do not benefit from adjuvant chemotherapy. At the same time a better understanding of tumour biology will also streamline the development of new regimens for those who are unlikely to benefit from existing drugs. It is expected that combinations of markers will be more informative to predict response than any single gene or gene product that may yield regimen-specific predictors. Novel molecular analytical tools, particularly transcriptional profiling, provide a method to test this hypothesis (7).

The use of cytotoxic chemotherapy in both advanced and early stage breast cancer have made significant progress in the last 10 years with several landmark studies identifying clear survival benefits for newer therapies. In spite of these developments the optimal approach for any specific patient can not be determined from a literature review or decision-making algorithm alone (3). Blanket application of published guidelines is usually unfeasible or inappropriate and careful consideration of the detailed circumstances of each patient is required to optimise the use of available treatment options.

2. Adjuvant therapy

Early breast cancer is disease that is confined to the breast alone or, in the case of women with node-positive disease, the breast and loco regional lymph nodes, and all detected disease can be removed surgically. With improved surgical techniques the delay in the initiation of chemotherapy post surgery is no longer an issue. Chemotherapy is routinely commenced within six weeks of surgery if indicated. However, micrometastatic disease may remain either locally or at distant sites that, if left untreated, could over the coming years develop into a life-threatening clinical recurrence.

Over the past few decades, many randomised trials have been undertaken of various treatments for early breast cancer,

but the duration of follow-up varies greatly between different trials and between different patients in the same trial. Hence, meta-analyses of the effects of such treatments on long-term outcome in various types of patient can deliver important insights into the value of different treatment concepts (5).

With continued improvements in local disease control (partly because of more careful and more extensive screening) and better use of systemic treatments both for early and for advanced disease a continued decrease in national mortality rates is anticipated.

Role of anthracyclines. Over the last thirty years, thousands of women have been enrolled into various clinical trials addressing questions over the role of chemotherapy versus no chemotherapy, role of polychemotherapy versus single agents, role of anthracyclines versus no anthracyclines, role of doses and schedules, and more recently adding taxanes and other novel compounds in chemotherapy arms. Progress has at times been pragmatic rather than logical, with new studies designed and initiated before the full results from previous trials have been available.

The evidence for systemic adjuvant chemotherapy for operable breast cancer originally came in 1968 from the NSABP conducted B-01 trial which investigated the role of thiotepa post-operatively (8). The NSABP (B-05) compared patients who received melphalan on 5 consecutive days every 6 weeks for 2 years with those who had been given placebo (9). Both studies showed significant improvement in the treatment arms. Another trial from Milan, investigating the combination of cyclophosphamide, methotrexate and 5-fluorouracil (CMF) every month for 1 year compared with an untreated group as adjuvant treatment to radical mastectomy in primary breast cancer with positive axillary lymph nodes showed a benefit for chemotherapy. After 27 months of study, relapse occurred in 24% of 179 control patients and in 5.3% of 207 women given combination chemotherapy [$P < 10^{-6}$], the advantage appeared statistically significant in all subgroups of patients. Patients with four or more positive axillary nodes had a higher number of relapses than those with fewer nodes (10). CMF was thus considered standard therapy for early breast cancer patients.

Since the mid-1980s, a number of randomised trials have investigated the role of anthracyclines. An absolute survival gain of 4% at 10 years is reported with the inclusion of anthracyclines (compared to non-anthracycline-based therapy) (5). The trials of combination chemotherapy with fluorouracil, adriamycin and cyclophosphamide (11) or fluorouracil, epirubicin and cyclophosphamide (FEC) versus no adjuvant chemotherapy yield breast cancer death rate ratios of 0.69 (SE 0.16) for younger and 0.79 (0.07) for older women; the trials of FAC or FEC versus CMF for 6-9 months yield ratios of 0.74 (0.06) for younger and 0.78 (0.08) for older women; and the trials of CMF alone for no more than 9 months versus no adjuvant chemotherapy yield ratios of 0.64 (SE 0.12) for younger and 0.93 (0.05) for older women. Combining these three meta-analyses yields the weighted averages of the breast cancer death rate ratios produced by FEC or FAC: 0.56 (SE 0.10, $2P < 0.00001$) for younger and 0.76 (0.06, $2P < 0.0001$) for older women. These results for about 6 months of FAC or FEC show clinically and statistically significant results, and



out as promising as the averaged results for all anthracycline-based regimens (of which the FAC or FEC results are a large part) (5).

Polychemotherapy using an anthracycline-containing regimen has been the cornerstone of treatment for women without pre-existing heart disease who require adjuvant chemotherapy for breast cancer (11-13). In the adjuvant setting, the French Adjuvant Study Group (FASG) initially defined the FEC 50 regimen as a reference treatment. The FASG-01 conducted in premenopausal, node-positive breast cancer patients showed that six cycles of FEC 50 were significantly better than three cycles of FEC 50 or FEC 75 in terms of 10-year DFS (14). The FEC regimen remains widely accepted in Europe, while four cycles of the AC regimen (doxorubicin and cyclophosphamide) is commonly used in the USA, based on the NSABP B-15 trial which showed results similar to those obtained using six cycles of CMF (cyclophosphamide, methotrexate, and fluorouracil) (15). AC was preferable since, following total mastectomy, AC was completed on day 63 versus day 154 for conventional CMF; patients visited health professionals three times as often for conventional CMF as for AC; and nausea-control medication was given for about 84 days to conventional CMF patients versus for about 12 days to patients on AC. On the other hand, when FEC regimens were compared with six cycles of CMF, the results were in favour of FEC, irrespective of epirubicin dose (50, 60, or 120 mg/m²) (Mouridsen H, *et al*, Proc ASCO 18: 1999; 16-18). Though in the study reported by Coombes *et al* using epirubicin 50 mg/m², there were two alternative schedules of FEC with FEC 1 having 50% lower dose intensity of cyclophosphamide and fluorouracil than FEC 2. There was a lack of benefit for FEC1 and overall there was no evidence of benefit for anthracycline containing FEC regimen over CMF. In the subgroup analysis of FEC2 versus CMF2, a modest but significant benefit was observed for the anthracycline containing regimen with an improved overall survival (P=0.02) and relapse free survival (P=0.03) (15). There are no phase III studies comparing epirubicin with doxorubicin as adjuvant therapy at optimal doses of each anthracycline. However, phase III comparisons of FEC and FAC at the same doses in MBC confirms similar efficacy, but with improved safety profile for epirubicin (18,19).

In the NEAT/SCTBG Br9601 trial (20) 2,391 pre- and post-menopausal, node positive and negative breast cancer patients were randomised to 8 cycles of CMF [classic CMF for NEAT patients, q3w i.v. CMF (cyclophosphamide = 750 mg/m², methotrexate = 50 mg/m², 5FU = 600 mg/m²) a for SCTBG patients] or 4 cycles of epirubicin (100 mg/m²) followed by 4 cycles of CMF (E→CMF) as above. After a follow-up of 32 months, a significantly better relapse-free survival (HR=0.70, P=0.0003) and OS (HR=0.64, P=0.0001) was seen in the anthracycline-containing arm. This 30% reduction in risk of relapse and 36% reduction in the risk of death with the sequential approach utilising E-CMF represents one of the largest differences seen in randomised comparisons of different chemotherapy regimens in breast cancer to date. The difference is almost twice as big as the improvement gained by the addition of 4 cycles of paclitaxel chemotherapy after 4 cycles of doxorubicin and cyclophosphamide (21).

The FASG-05 study showed that fluorouracil 500 mg/m², cyclophosphamide 500 mg/m², and epirubicin 100 mg/m² (FEC 100) was better than the same regimen with epirubicin 50 mg/m² (FEC 50) in terms of disease-free survival (DFS) and overall survival (OS) in adjuvant treatment of early breast cancer (22). In this study, 565 patients were randomised to treatment with FEC 50 or FEC 100 after surgery. Post-menopausal patients also received tamoxifen for 3 years. The 10-year DFS was 45.3% (95% CI, 41.9-48.7%) with FEC 50 and 50.7% (95% CI, 47.3-54.1%) with FEC 100 (P=0.08). The 10-year OS was 50.0% (95% CI, 46.7-53.3%) with FEC 50 and 54.8% (95% CI, 51.3-58.3%) with FEC 100 (P=0.05). Delayed cardiac toxicity (before relapse) occurred in four patients (1.5%) in the FEC 50 arm and three patients (1.1%) in the FEC 100 arm (23). The authors concluded that treatment with adjuvant FEC 100 demonstrated superior DFS and OS versus FEC 50 at 10 years of follow-up. The long-term safety of the FEC regimen that was demonstrated at 5 years was maintained at the 10-year analysis. There was no significant difference in the rate of delayed cardiac toxicity between the FEC 100 and the FEC 50 group. Given the risk-benefit ratio, FEC 100 is a more optimal regimen for long-term survival in patients with poor prognosis. It is interesting here to note that there is no doxorubicin dose effect as was shown in the study where multiple doses (60/75/90 mg/m²) of doxorubicin did not show any differences in response (21). Based on the evidence discussed above, anthracyclines are now considered standard adjuvant therapy for patients with high-risk early breast cancer.

Role of taxanes. Taxanes have emerged as critically important drugs in the treatment of breast cancer. Development of these agents is characterised by rapid collection of an unprecedented amount of data from numerous large, high-quality prospective random assignment trials that involved tens of thousands of patients (24).

Five trials of adjuvant chemotherapy compared a taxane-containing regimen with a non-taxane containing regimen. Involving more than 9,000 women with 2,512 relapses and 1,591 deaths the treatment approaches that were investigated were heterogeneous. Three trials used paclitaxel (Mamounas EP, *et al*, Proc ASCO 22: 2003; 21,25) and two used docetaxel (Martin M, *et al*, San Antonio Breast Cancer Symposium, abs. 43, 2003; Jones SE, *et al*, Proc ASCO 22, 2003). Paclitaxel 3-h infusion was used in two trials (21) and 24-h infusion in one trial (25). Although all trials used a 21-day schedule in the taxane-containing group, the total number of cycles of chemotherapy ranged from four to eight. Three trials (25) used the same numbers of cycles of chemotherapy in both groups, whereas two trials (21), used twice as many cycles in the taxane group (increasing from four to eight cycles in both studies), confounding the use of a taxane with the addition of more cycles of chemotherapy. This has drawn criticism to the trial design because the regimens were of different durations and suggested that the favourable results in the AC/taxane arm may be due to the delivery of additional cycles of chemotherapy rather than a distinct taxane benefit. In CALGB 9344 study, 3,121 women with operable node positive breast cancer were randomly assigned post-operatively to receive a combination of cyclophosphamide

(C), 600 mg/m², with one of three doses of doxorubicin (A), 60, 75, or 90 mg/m², for four cycles followed by either no further therapy or four cycles of paclitaxel at 175 mg/m². Tamoxifen was given to 94% of patients with hormone receptor-positive tumours. There was no evidence of a doxorubicin dose effect. At 5 years, disease-free survival was 69, 66, and 67% for patients randomly assigned to 60, 75, and 90 mg/m² of doxorubicin, respectively. The hazard reductions from adding paclitaxel to CA were 17% for recurrence and 18% for death. At 5 years, the disease-free survival (\pm SE) was 65% (\pm 1) and 70% (\pm 1), and overall survival was 77% (\pm 1) and 80% (\pm 1) after CA alone or CA plus paclitaxel, respectively. The effects of adding paclitaxel were not significantly different in subsets defined by the protocol, but in an unplanned subset analysis, the hazard ratio of CA plus paclitaxel versus CA alone was 0.72 (95% confidence interval, 0.59-0.86) for those with oestrogen receptor-negative tumours and only 0.91 (95% confidence interval, 0.78-1.07) for patients with oestrogen receptor-positive tumours, almost all of whom received adjuvant tamoxifen. The additional toxicity from adding four cycles of paclitaxel was generally modest (21). In another published prospective randomised trial, the role of paclitaxel was evaluated in an adjuvant setting to determine its impact on reducing the risk of recurrence in patients with operable breast cancer. Five hundred and twenty-four patients were randomised to be treated either with 4 cycles of paclitaxel followed by 4 cycles of combination therapy with FAC (Pac/FAC) or with 8 cycles of FAC alone. Patients with intact primary breast cancer received the initial 4 cycles of paclitaxel or 4 cycles of FAC in a neoadjuvant setting. After completion of 8 cycles of chemotherapy, those patients who were \geq 50 years and whose tumours were positive for oestrogen receptors received tamoxifen for 5 years. Estimated DFS at 48 months was 0.83 for FAC and 0.86 for Pac/FAC group. The overall estimated hazard ratio for Pac/FAC compared with FAC derived by fitting the Cox regression model and incorporating terms for prognostic factors was 0.66. Preliminary results suggest that the addition of paclitaxel to a FAC regimen of adjuvant or neoadjuvant therapy may further reduce the risk of disease recurrence; however, differences were not statistically significant ($P=0.09$). The survival data are too preliminary to permit meaningful evaluation of the impact of paclitaxel on mortality (25).

In the BCIRG 001 trial, 1,491 patients were randomised to receive treatment with either TAC (docetaxel/doxorubicin/cyclophosphamide at doses of 75/50/500 mg/m², respectively) or FAC (at doses of 500/50/500 mg/m², respectively) every 3 weeks for six cycles. Both treatments were well tolerated, with high rates of treatment compliance and completion. The TAC regimen was found to be superior to FAC in terms of the primary end point, DFS. TAC produced a 28-32% reduction in the risk of recurrence in the two analyses. The higher DFS rate in the TAC group translated into a survival benefit; the initial trend toward longer survival with TAC at 33 months reached statistical significance at the 55-month median follow-up.

A systematic review of randomised trials of adjuvant or neoadjuvant systemic therapy recently published identified ten reported trials comparing a taxane-containing group with

a non-taxane-containing control group in women with early breast cancer (26). Four of five neoadjuvant trials showed higher rates of complete response with taxanes, although differences were not significant. All five adjuvant trials showed improvements in DFS with taxanes, and these improvements were significant in three trials and were independent of oestrogen-receptor status. Two trials showed a significant improvement in overall survival. These results are used to support the use of adjuvant taxanes in women with early node positive breast cancer. Longer follow-up of these trials and results from ongoing trials are needed to clarify the best use of taxanes in early breast cancer. The strongest evidence is for the addition of four cycles of paclitaxel to four cycles of doxorubicin and cyclophosphamide, or for the substitution of six cycles of FAC with six cycles of docetaxel, doxorubicin, and cyclophosphamide. This effect is independent of hormone-receptor status, and the evidence does not support restricting the use of taxanes to women with hormone-receptor negative tumours. There is also evidence suggesting that docetaxel and cyclophosphamide is an acceptable alternative to doxorubicin and cyclophosphamide, although long-term data on this regimen is lacking. Roche *et al* presented the data on the 6 cycles of FEC 100 (Arm A) as compared to 3 cycles of FEC 100 followed by 3 cycles of Docetaxel (Arm B) for node positive breast cancer patients. Between June 1997 and March 2000, 1,999 patients were recruited in 83 French and Belgian centres. More febrile neutropenia and nail disorders were observed in Arm B and a more decreased and subnormal LVEF at the end of chemotherapy in Arm A. Five cases of leukaemia (3 Arm A; 2 Arm B) were observed. No toxic deaths have been reported. The substitution of 3 cycles of docetaxel for 3 cycles of FEC 100 following 3 cycles of FEC 100 significantly improved DFS and overall survival. There was a 17% reduction in the risk of relapse (HR 0.83, range 0.69-0.99) with a P-value of 0.041. Five-year overall survival was 90.7% for the sequential arm as compared to 86.7% for 6 cycles of FEC 100 arm ($P=0.017$). There was thus a 23% reduction in the risk of death (HR 0.77, range 0.59-1.00) with a P-value of 0.05 (Roché H, *et al*, San Antonio Breast Cancer Symposium 2004).

There are 15 unreported trials including more than 19,000 women addressing similar and related questions (Riou JF, *et al*, Proc AACR 385: 1994). Longer follow-up of all trials is needed to clarify the role of taxanes in the treatment of early breast cancer. Based on indirect comparisons as well as the results of the recent randomised trial conducted in patients with MBC, docetaxel appears to be the more active taxane. In addition to its longer half-life, docetaxel also has a more rapid cellular uptake and longer intracellular retention than paclitaxel. Because of its pharmacokinetics, the efficacy of paclitaxel is schedule-dependent. In general, trends of superior response rates have been associated with higher doses and prolonged infusions times, but no regimen of paclitaxel has been shown to be statistically superior to any other in MBC. Docetaxel is highly active when given as a short, intermittent infusion. Dose-dense paclitaxel-based therapy, in which chemotherapy cycles are administered every 2 weeks has produced impressive results in the adjuvant setting. It remains to be determined if this approach is superior to conventional docetaxel-based therapy in this setting or if a



SPANDIDOS PUBLICATIONS docetaxel-based regimen will be similarly. At the current time, clinicians should choose a taxane-based regimen for their patients with breast cancer based on consideration of the pharmacokinetics, clinical activity, toxicity and dosing schedule that best meets the patients' needs (24). Results from ongoing and recently completed trials will no doubt improve outcome and quality of life for patients with early-stage breast cancer. Future trials should shed light on their ideal amalgamation with existing and promising treatments.

Trastuzumab in adjuvant setting. Trastuzumab is a rationally designed compound that targets cancer cells that overexpress HER-2 cell surface signal transduction protein. About 20% of breast cancers overexpress HER-2 as a result of gene amplification. Stimulation of HER-2 is associated with increased cell growth, angiogenesis and inhibition of apoptotic signals.

The National Surgical Adjuvant Breast and Bowel Project (NSABP) study and the North Central Cancer Treatment Group (NCCTG) study performed a joint interim analysis which involved 3,351 breast cancer patients treated with adjuvant trastuzumab.

Both randomised controlled trials evaluated the combination of anthracycline and cyclophosphamide (AC) followed by paclitaxel chemotherapy, with or without Herceptin in women with HER-2-positive breast cancer. Results showed that patients who received trastuzumab in combination with standard combination chemotherapy had a 53% reduction in risk of disease recurrence compared to those treated with chemotherapy alone. There were 96 events in the Herceptin arm as compared to 191 events in the non-herceptin arm with a hazard ratio of 0.47 and a highly significant P-value of 8×10^{-10} (27). Subsequently the Breast International Group (BIG) reported an analysis of the HERA (Herceptin Adjuvant) study. This large, phase III randomised controlled trial for patients with early stage HER2-positive breast cancer involved nearly 5,100 patients enrolled at 480 sites in 39 countries worldwide. The interim analysis compared 12 months of herceptin versus observation and did not include a comparison of 24 months of herceptin versus observation. These data will become available as the study matures. There was a significant improvement in the 2-year DFS with the addition of 1-year trastuzumab ($P=0.0001$). The 2-year DFS improved from 77.4% in the observation arm to 85.8% in the 1-year trastuzumab arm (HR 0.54; 95% CI 0.43-0.67) (28). These results had huge implications in management of early HER-2 positive breast cancer. Herceptin is now the standard of care in HER-2 positive breast cancer patients. These results confirmed the notion that a better understanding of the biology of the tumour and pathways of cell proliferation will help in the development of new treatments directed against the same processes that lead cancer cells to evade apoptosis. This is a big step forward for thousands of women with breast cancer and we now have an effective weapon against HER-2 positive disease by targeting HER 2 positive cancer cells.

3. Chemotherapy for metastatic breast cancer (MBC)

Although generally incurable, MBC remains chemosensitive such that symptom palliation and prolongation of survival

can be achieved. However, response duration remains disappointingly short- and long-term survival remains uncommon, such that ongoing research is required.

Anthracyclines. Anthracyclines possess significant activity in chemo-naïve patients or those who received them in the adjuvant setting more than 12 months ago. Response rates of 30-40% have been documented in patients with MBC (29,30). Despite their significant role in the adjuvant setting, the use of anthracyclines in patients with MBC may be limited by significant toxicity.

Meta-analysis of 30 trials has shown that polychemotherapy regimens containing anthracycline were associated with superior response rate, but without a significant survival benefit compared to regimens without anthracycline, and with increased gastro-intestinal toxicity, cardiotoxicity and alopecia (31). This meta-analysis is, however, limited by its use of published data only as well as the heterogeneity of patients and their previous treatments. Furthermore, analysis suggested that the addition of an anthracycline to a chemotherapeutic regimen did in fact improve overall survival (32).

PEGylated liposomal doxorubicin may result in improved pharmacokinetics and preferential accumulation of drug in the tumour. Such tumour selectivity is thought to be mediated by increased permeability of tumour vasculature and impaired lymphatic drainage allowing accumulation of macromolecules, the so-called enhanced permeability and retention effect (33).

Taxanes. In the early 1990s paclitaxel was identified as an agent with significant activity against MBC with objective response rates in excess of 50% (34,35). Subsequently, the semi-synthetic taxane docetaxel was also found to be an active drug in this setting.

For patients exposed to anthracyclines in the adjuvant setting or who have failed in the metastatic setting, taxane-based treatment is currently the standard of care. This is based on a phase III randomised controlled trial comparing single agent docetaxel with mitomycin plus vinblastine (36). Overall survival was 11.4 versus 8.7 months favouring the docetaxel arm ($P=0.0097$). Response rate (30 versus 12%) and TTP (4.4 versus 2.5 months) were also statistically superior in the taxane group. Similarly, docetaxel has demonstrated superiority over combination chemotherapy comprising methotrexate plus 5-FU in terms of response rate (42 versus 21%) and TTP (6.3 versus 3 months). There was no statistically significant difference in overall survival in this study, although there was cross-over upon progression which may account for this difference (37). Similarly, paclitaxel has compared favourably with CMF plus prednisolone when used as 1st line therapy for MBC (38), although taxol was inferior to the combination of cisplatin with etoposide (39).

Taxanes in anthracycline naïve patients. For patients not previously exposed to anthracyclines, taxanes have been directly compared to anthracycline in randomised trials. In one study of 326 patients, patients receiving docetaxel (100 mg/m² q21 days) achieved an objective response rate of 48% compared to 33% in patients treated with doxorubicin

(75 mg/m²). Docetaxel was also associated with a trend to improved TTP (26 versus 21 weeks; $P=NS$) and reduced myelotoxicity, but there was no difference in overall survival (15 versus 14 months) (29).

Conversely, paclitaxel (200 mg/m² over 3 h every 21 days) has been shown to be inferior to doxorubicin (75 mg/m²). In a trial of 331 patients paclitaxel showed inferior response rate (25 versus 41%; $P=0.003$), inferior PFS (3.9 versus 7.5 months; $P<0.001$), and a trend towards inferior survival (15.6 versus 18.3 months) (30). In a separate study paclitaxel administered as a 24-h infusion was equivalent to doxorubicin, although this may be attributable to the lower dose of doxorubicin used in this study (60 mg/m²) (40).

The inference from these data is that docetaxel may be superior to paclitaxel in MBC. Indeed, a trial directly comparing the two confirmed superior TTP and overall survival favouring docetaxel, but with a higher incidence of myelosuppression (41).

Taxane scheduling. Further investigations have attempted to define the optimal schedule for taxane administration. A randomised phase II study comparing docetaxel given weekly versus 3-weekly demonstrated significantly less haematological, neurological and gastrointestinal toxicity (although paradoxically more patients withdrew from the weekly arm due to toxicity), whilst response rates were identical (34 versus 33%) (42). Of note, higher doses of steroid were used in the weekly arm, which may account for some of the decreased toxicity.

Similarly, in a study of 585 patients comparing paclitaxel given weekly or 3-weekly favoured the weekly schedule in terms of response rate (40 versus 28%; $P=0.017$) and TTP (9 versus 5 months; $P=0.0008$) with a non-significant trend to improved survival (24 versus 16 months) (43). The weekly schedule was associated with less haematological toxicity, although neurotoxicity was greater. These data suggest that the apparent inferiority of paclitaxel in previous phase III studies may have been due to use of a sub-optimal schedule.

Taxane-based combination therapy. Despite encouraging phase II data for the combination of taxane with anthracycline, disappointingly, the substitution of docetaxel for 5-FU in combination with doxorubicin and cyclophosphamide did not improve TTP or survival when used as 1st line treatment in MBC (Mackey JR, *et al*, Proc ASCO 137: 2002). Similarly taxane-anthracycline combinations have failed to show significant benefit compared to non-taxane containing combinations in other randomised phase III trials. A median number of six cycles were delivered in the two treatment arms. Dose escalation was only possible in 17 and 20% of the AT and AC patients, respectively. Median PFS was 6 months in the two treatments arms. Response rate was 58 versus 54%, and median overall survival was 20.6 versus 20.5 months in the AT and AC arms, respectively. The AT regimen was characterized by a higher incidence of febrile neutropenia (32 versus 9% in the AC arm). No differences in the efficacy study end points were observed between the two treatment arms. Treatment-related toxicity compromised doxorubicin-delivered dose-intensity in the paclitaxel-based regimen (44). This is in contrast to the positive effect of the

addition of taxane to anthracycline in the adjuvant setting (Nabholtz JM, *et al*, Proc ASCO 141a: 2002). In the meta-static setting, the data may at least in part, be confounded by the use of the experimental treatments as a salvage therapy for patients initially assigned to the control arm of the study.

Data pertaining to the combination of paclitaxel with anthracyclines have also been generally disappointing when compared to non-taxane containing doxorubicin-based regimens. Overall response rates for patients randomized to AT and FAC were 68 and 55%, respectively ($P=0.032$). Median TTP and overall survival were significantly longer for AT compared with FAC [time to progression 8.3 months versus 6.2 months ($P=0.034$); overall survival 23.3 months versus 18.3 months ($P=0.013$)]. Therapy was generally well-tolerated (median of eight cycles delivered in each arm). Grade 3 or 4 neutropenia was more common with AT than with FAC (89 versus 65%; $P<0.001$); however, the incidence of fever and infection was low. Grade 3 or 4 arthralgia and myalgia, peripheral neuropathy, and diarrhoea were more common with AT, whereas nausea and vomiting were more common with FAC. The incidence of cardiotoxicity was low in both arms. Interestingly only one-quarter of patients assigned FAC subsequently received a taxane in this study (45). However, the addition of paclitaxel to doxorubicin may increase cardiotoxicity perhaps due to paclitaxel interactions resulting in higher levels of cardiotoxic doxorubicin metabolites, an effect not observed with docetaxel (46).

The generally disappointing results of taxane-anthracycline combinations in MBC may not be altogether surprising since there is no compelling pre-clinical data indicating synergy between these classes of drugs and the primary toxicity for both is haematological.

4. Other chemotherapeutic agents with activity in MBC

Capecitabine is an oral fluoropyrimidine prodrug activated by thymidine phosphorylase, which affords a degree of tumour selectivity since this enzyme is preferentially expressed in tumour tissue (47). Capecitabine has demonstrated single agent activity in MBC, with a response rate in excess of 20% and median survival greater than 1 year even in patients with disease refractory to both anthracycline and taxane, and with a favourable toxicity profile and oral bioavailability (48).

In phase II studies, vinorelbine has achieved response rates of up to 50% in the setting of MBC and was superior to melphalan in an RCT in patients with anthracycline-refractory disease. TTP was significantly longer with vinorelbine than with melphalan (median TTP 12 weeks versus 8 weeks, respectively ($P<0.001$)). The effect of vinorelbine on survival was also statistically significant ($P=0.034$). The 1-year survival rates were 35.7 versus 21.7% in favour of vinorelbine and median survival rate was 35 and 31 weeks, respectively. In total, 46.5% of vinorelbine patients and 28.2% of melphalan patients achieved an objective response or stabilization of disease ($P=0.06$). The most common toxicities were hematologic, including granulocytopenia with vinorelbine and thrombocytopenia and granulocytopenia with melphalan. Both drugs were generally well tolerated with no reported treatment-related deaths (49).



II studies have confirmed activity of gemcitabine in response rates of up to 37% as first line therapy (50-51). However, no significant activity has been reported in patients with anthracycline or taxane-refractory disease. The only phase II study in this setting did not demonstrate any objective response (52).

The degree of activity and favourable toxicity profiles of all these drugs has lead to their investigation earlier in the course of MBC and in combination with other agents.

Chemotherapy in combination with trastuzumab. Trastuzumab in combination with chemotherapy has demonstrated a survival benefit over chemotherapy alone in patients with Her-2/neu expressing breast cancer.

Trastuzumab in combination with either doxorubicin and cyclophosphamide or with paclitaxel achieved significantly greater TTP, response rates, and 2-year survival compared to chemotherapy alone (53).

In support of these clinical observations, *in vitro* data suggests additive or even synergistic interaction between trastuzumab and chemotherapeutic agents including taxanes and also platinum agents (54). These data have prompted phase II trials of trastuzumab with either docetaxel/cisplatin or docetaxel/carboplatin in MBC with response rates of RR of 64-76% (Slamon DJ, *et al*, Proc ASCO 193a: 2001; Pienkowski T, *et al*, Proc ASCO 2030a: 2001). These data confirm the potential for trastuzumab to enhance chemosensitivity and underpin its use in the adjuvant setting where it has recently been confirmed to have a significant impact.

5. Combination versus sequential single agent chemotherapy in MBC

Based on the success of combination chemotherapy in curing lymphoma and germ cell tumours and the theoretical principles of non-cross resistant drugs with non-overlapping toxicities, it is becoming increasingly common to use combination chemotherapy in most tumour sites. It has been commonly assumed that combination chemotherapy for MBC will result in improved response rate, better symptom palliation and improved survival compared to single agent chemotherapy. However, this may only be true for drug combinations with true synergistic potential. These principles were supported by data from randomised controlled trials and a metaanalysis in the pre-taxane era with polychemotherapy resulting in superior response rates and overall survival compared to single agent. However, these trials did not directly address the question of combination therapy versus sequential monotherapy with the same agents. Since it is also commonly held that anthracyclines and taxanes are the most active single agents against MBC, the combination of these two classes of drug holds great interest (3).

Controversy remains over the use of combination versus sequential single agent therapy with no combination previously demonstrating superiority in terms of overall survival compared to sequential use of the same agents. A seminal study addressed this question by comparing anthracycline followed by taxane with taxane followed by anthracycline and with anthracycline plus taxane in combination. There was no significant difference in the two sequential arms of this study.

Despite improved response rate (doxorubicin 36%; paclitaxel 34%; combination 47%) and TTP (doxorubicin 5.8 months; paclitaxel 6 months; combination 8 months) in favour of the combination arm, this did not translate in to a significant survival advantage over sequential therapy (doxorubicin 18.9 months; paclitaxel 22.2 months; combination 22 months). Furthermore, combination therapy was not associated with any improvement in quality of life (40). Reasons for the failure of combination therapy in this study may include the lack of true synergy between anthracyclines and taxanes, and the predominant toxicity of both drugs being haematological such that their combination may compromise drug dosage. Such limitations may be overcome by the rational use of drug combinations based on true synergy in pre-clinical models (3).

Pre-clinical studies have indicated that taxanes can upregulate thymidine phosphorylase, the activating enzyme of capecitabine. Further, synergistic interaction of taxane plus capecitabine has been observed *in vivo* xenograft models (55,56). This underpins the rationale for the combination of docetaxel with capecitabine in clinical trials.

In a phase III trial of 511 women with MBC who had received a prior anthracycline regimen patients were randomised to receive either docetaxel (100 mg/m² q21 days) or the combination of docetaxel (75 mg/m² q21 days) plus capecitabine (57). Despite the lower dose of docetaxel, the combination had a higher response rate (42 versus 30%, $P=0.006$), improved TTP (6.1 versus 4.2 months; $P=0.0001$), and longer overall survival (14.5 versus 11.5 months; $P=0.0126$). This was at the expense of greater toxicity, with 65% of patients in the combination arm requiring dose modification compared to 36% in the docetaxel only arm. Although a large number of patients in both groups received subsequent chemotherapy upon progression, a major criticism of this study is the low use of subsequent capecitabine in patients originally assigned to the docetaxel only arm, with only 27% of patients receiving capecitabine after progression on docetaxel. Interestingly, those that did go on to receive capecitabine appeared to have a survival advantage over patients receiving other salvage chemotherapy. Thus, although this trial demonstrated superiority in terms of response rate, TTP and overall survival for docetaxel plus capecitabine compared to single agent docetaxel, it did not address the question of combination treatment versus sequential use of docetaxel followed by capecitabine. This remains a question of great interest.

In summary, although studies have indicated that response rates and TTP may be improved by combination treatment, in those where the same agents have been given sequentially, the overall survival is equivalent. Nevertheless, combination therapy may be more appropriate where a higher response rate is important, for example in the neo-adjuvant setting or in the presence of bulky visceral metastases, and also potentially in the adjuvant setting. In this latter respect, trials of combination chemotherapy in MBC may remain the test bed for potentially more active new combinations for translation to the adjuvant setting. This is exemplified by the comparison of TAC versus FAC in MBC where the substitution of 5-FU by docetaxel to the combination of doxorubicin and cyclophosphamide did improve response

rate but not overall survival. However, in testing the same regimens in the adjuvant setting there was a clear improvement in survival in the taxane containing arm. Furthermore, there remains a risk with sequential therapies that some patients may not go on to receive all active drugs, presumably due to deterioration in performance status or development of organ dysfunction precluding the use of further chemotherapy (3). This is demonstrated in the capecitabine/docetaxel study in which a third of patients in the docetaxel only arm did not receive any subsequent chemotherapy.

In conclusion, there is no clear evidence that one agent or combination of agents is superior to another in the management of MBC and treatment will be increasingly influenced by what has been used in the adjuvant setting and the treatment free interval, toxicity profile, mode of administration, and patient choice. Apart from Her-2 there are currently no molecular markers to influence the choice of chemotherapy for individual patients and translational end points with which to guide this should increasingly be incorporated into future trials of chemotherapy for MBC so that treatment may consist of individually tailored regimens based on biological prognostic markers.

Receptor status and use of taxanes: what do we know? A review of the data on individual patients from the BCIRG 001 and PACS 01 trials were combined to examine the effect of hormone receptor status on taxane efficacy (58). Hazard ratios for recurrence and survival were estimated by Cox proportional hazards models and were adjusted for clinical variables. Interaction between docetaxel and ER expression was tested. ER status was available for 3,329 patients (95% of all randomly assigned patients), of whom 75% (n=2,493) were ER positive. Docetaxel therapy was associated with a 30% reduction in the risk of death [hazard ratio (HR)=0.70; 95% CI, 0.54 to 0.91] in ER-positive patients and a 31% reduction (HR=0.69; 95% CI, 0.52-0.94) in ER-negative patients. Docetaxel therapy was associated with a 21% reduction in the risk of recurrence (HR=0.79; 95% CI, 0.66-0.93) in ER-positive patients and a 31% reduction (HR=0.69; 95% CI, 0.54-0.97) in ER-negative patients. The interaction between docetaxel therapy and ER status was not statistically significant for either overall survival (P=0.87) or DFS (P=0.30). ER expression was also not predictive for docetaxel efficacy when it was analyzed as a semi-continuous variable based on percent of positive cells by immunohistochemistry. The issue of whether HER2 status is a predictive marker of benefit from taxane therapy is particularly contentious. A retrospective analysis of a subset of patients in the CALGB 9344 study suggested that the addition of paclitaxel to AC was only beneficial for women with early-stage breast cancer whose tumours overexpressed HER2, regardless of their ER status (59). This study randomly selected 1,500 women from 3,121 women with node-positive breast cancer who had been randomly assigned to receive doxorubicin plus cyclophosphamide for four cycles, followed by four cycles of paclitaxel or observation. Tissue blocks from 1,322 of these 1,500 women were available. Immunohistochemical analyses of these tissue specimens for HER2 with the CB11 monoclonal antibody against HER2 or with a polyclonal-antibody assay kit and fluorescence *in situ* hybridization for HER2 amplification were

performed. The interaction between HER2 positivity and the addition of paclitaxel to the treatment was associated with a hazard ratio for recurrence of 0.59 (P=0.01). Patients with a HER2-positive breast cancer benefited from paclitaxel, regardless of estrogen-receptor status, but paclitaxel did not benefit patients with HER2-negative, estrogen-receptor-positive cancers. Likewise, a subgroup analysis of the UK TACT trial suggested that patients with tumours that over-express HER2 may derive benefit from the addition of a taxane, although this benefit was confined to the HER2-positive tumour subgroup with absent hormonal receptor expression. Immunohistochemical staining of tumour samples from the GEICAM 9906 study also suggested that the addition of paclitaxel was only associated with improved DFS in patients whose tumours lacked HER2 expression (Rodríguez-Lescure A, *et al*, J Clin Oncol 25 (Suppl.): a10598, 2007). Patients were randomly assigned to receive either four cycles of standard-dose AC; n=510, or TC; n=506, administered by intravenous infusion every 3 weeks. Baseline characteristics in the two age subgroups were generally well matched, except that older women tended to have more lymph node involvement. At a median of 7 years follow-up, the difference in DFS between TC and AC was significant (81% TC versus 75% AC; P=0.033; hazard ratio, 0.74; 95% CI 0.56-0.98) as was OS (87% TC versus 82% AC; P=0.032; HR, 0.69; 95% CI, 0.50-0.97). TC was superior in older patients as well as younger patients. There was no interaction of hormone-receptor status or HER2 status and treatment. Limited data from the HeCOG 10/97 study and the USO 9735 trial also failed to demonstrate that the benefit of taxane administration was confined to patients with HER2-positive disease (60,61). Based upon these findings, the 2007 ASCO guidelines do not support the use of HER2 expression as a marker to guide decisions on taxane administration in the adjuvant setting (62). Determination of ER or HER2 status alone is unlikely to reveal which patients are likely to benefit from the inclusion of a taxane as adjuvant therapy. To date, none of the first-generation taxane trials has assessed whether a combination of ER, HER2, and proliferative markers can predict taxane efficacy across the molecular subgroups. In the PACS 01 trial, examination of a panel of immunohistochemical tissue microarray markers suggested that the addition of docetaxel was most beneficial in the basal-like subgroup of breast cancers, marked by the absence of ER, PR and HER2 expression (Jacquemier J, *et al*, J Clin Oncol 24: a509, 2006). Further validation of such preliminary evidence is eagerly awaited from other first-generation taxane trials.

In the future, the incorporation of novel biomarkers into clinical trial designs, combined with improved classification of breast cancer molecular subtypes, may help to predict whether individual patients are likely to benefit from taxane treatment. Taxanes exert their cytotoxic effects in the G2/M phase of the cell cycle by inhibiting microtubule disassembly, which retards mitotic activity in malignant cells. A variety of potential mechanisms can lead to taxane resistance. Over-expression of P-glycoprotein, a transmembrane efflux pump, is thought to be a common mechanism of taxane resistance (63,64). Compared with Pgp-negative tumours, a significant increase in doxorubicin and Taxol resistance was seen for breast cancers that expressed Pgp, regardless of prior treat-



strong correlation between the degree of Pgp expression and *in vitro* resistance to Taxol and doxorubicin (but not to 5-fluorouracil) was found when either IHC scores or image analysis-based methods were used to quantify Pgp expression ($n=185$, $P<0.0001$). The degree of Pgp expression strongly correlated with the degree of drug resistance in the clinical specimens studied. These data suggested that Pgp contributes to clinical MDR1-related drug resistance, and both intrinsic and acquired expression of Pgp in breast cancer may contribute in part to therapeutic failure and relapse. A meta-analysis of breast cancer trials indicates that this glycoprotein is expressed in 41% of tumours and that its expression increases with exposure to chemotherapy (65). However, clinical data on taxane sensitivity are scarce and early-phase clinical trials of P-glycoprotein inhibitors have been disappointing (66).

Based upon the first-generation taxane trials, taxane-based regimens are an important addition to the armamentarium against early-stage breast cancer. However, deciding which individual patients are likely to benefit from taxanes remains a challenge. Ultimately, clarifying the role of factors that affect the efficacy of taxanes in particular subgroups of patients (defined by traditional biomarkers such as age, ER status, and HER2 expression) will require international collaboration with rigorous examination of data from individual patients in first-generation taxane trials, in a future EBCTCG meta-analysis (67). Validation of additional biomarkers and evaluation of novel agents in the adjuvant setting will require innovative approaches to clinical trial design. Randomised clinical trials with built in translational studies with commitment to novel platforms for translational research will help improve the understanding of tumour biology thus moving us towards an era of rationally tailored therapy in the near future.

6. Conclusion

Increasing evidence suggests we need to apply different approaches to understanding the role of chemotherapy in ER-positive and ER-negative disease. ER-negative disease tends to have a disproportionate impact on early events in trials of adjuvant chemotherapy partly as a result of being a generally more aggressive phenotype and partly because the impact of adjuvant endocrine therapy suppresses early relapses in ER-positive disease. Recent research has allowed us to refine breast cancers further into prognostic groups based on gene expression profile. It is anticipated that we will be able to utilise expression profiles to guide adjuvant chemotherapy decisions in patients where the indication for chemotherapy on conventional grounds is borderline, and to avoid unnecessary chemotherapy in patients with adverse conventional features who carry a favourable gene expression signature since benefit in these patients is predicted to be low. Clinical trials to prove the value of this approach are currently being designed. Assuming these approaches are successful we will need to develop our ability to roll out the findings into routine practice. While there remains much potential for further refinement of conventional cytotoxic agents the largest leaps forward are likely to come from incorporation of targeted therapies. Trastuzumab has produced quite startling results in the adjuvant context and whilst unrealistic to expect many more

results of this magnitude new agents such as the VEGF antibody bevacizumab are now showing promising results in advanced disease in breast cancer, and are being tested in the adjuvant setting in high risk patient groups. The future for breast cancer therapy is brimming with promise but it is important to be prepared for disappointment and remember that no amount of mice cured in model experiments can guarantee a successful human therapy. Breast cancer therapy development will however probably remain true to form of the last 30 years with drugs demonstrating clear superiority in controlling advanced disease proving to be even more valuable in treatment of early disease. The abundance of molecular therapies that are emerging from better understanding of cancer biology provides an optimistic climate for the future of breast cancer.

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