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Case histories: breast cancer

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1. Introduction

Recent years have seen both the introduction of a number of new targeted therapies, which have transformed the management of breast cancer, and a growing understanding of how existing treatments such as anthracyclines can be optimised. The potential role of these new treatment options is illustrated in the following case histories. It should be noted that these histories were developed to generate discussion at the workshop and do not describe actual cases, but combine elements from several patients to highlight the various challenges faced by clinicians.

2. Triple-negative breast cancer

In August 2004, an asymptomatic, nulliparous, 29-year-old woman noticed a palpable left breast nodule. Her mother had previously experienced breast cancer, and her maternal grandmother had experienced both breast and ovarian cancer. Performance status assessed according to the Eastern Cooperative Oncology Group (ECOG) was 0.

Early-onset breast cancer and breast/ovarian cancer in close relatives from the same side of the family are two criteria for an increased risk of hereditary breast cancer. Women who have inherited mutations in the *BRCA1* and/or *BRCA2* genes have a high risk of developing breast and/or ovarian cancer. Breast cancer survivors with an inherited mutation in either gene are at increased risk of developing a second primary breast cancer. [1] In addition, women with mutations in these genes are at markedly increased risk of other cancers, such as ovarian cancer, and often receive a cancer diagnosis earlier than women in the general population. [1]

We decided to ask this patient to consider genetic testing for BRCA1 and BRCA2 mutations. The patient accepted and was found to have a positive BRCA1 test result.

Bilateral mammography and breast ultrasound revealed a 2.5 cm hypoechoic area, with irregular borders, in the left upper external quadrant, and two mobile round nodes in the left axilla. Nodule core biopsy showed ductal infiltrating carcinoma (cT2cN1); the tumour was negative for oestrogen receptors (ER), progesterone receptors (PR) and human epidermal growth factor receptor-2 (c-erbB2), and Ki-67 proliferative index expression was 80%.

This profile is typical of the triple-negative subtype of breast cancer. Compared with other phenotypes, triple-negative breast cancer is associated with an

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increased risk of distant recurrence during the first 3–4 years after diagnosis, an increased risk of visceral and brain metastases, and a poor prognosis; in one study, the median survival after recurrence was 9 months, compared with 20 months in patients with other phenotypes.[2,3] Young women (<35 years) with triple-negative breast cancer have a worse prognosis than older women with similar disease features.[4] Aggressive first-line treatment is therefore warranted in such patients.

Neoadjuvant chemotherapy appears to produce higher pathological complete response rates in patients with triple-negative breast cancer than in those with other disease phenotypes; the greatest benefits are achieved with sequential anthracycline and taxane regimens.[5] Such regimens produce good survival rates in patients in whom pathological complete responses are achieved. However, in cases of residual disease, survival rates are lower in patients with triple-negative disease than in patients with other phenotypes.[6] In phase II trials, neoadjuvant therapy with cisplatin-based regimens has also been shown to be effective in patients with triple-negative breast cancer.[7–9] A recent study has suggested that decreased *BRCA1* expression may be predictive of response to platinum-based therapy in these patients.[7]

Based on this patient's preference, we decided to avoid neoadjuvant treatment, instead recommending the patient for surgery. The patient underwent left quadrantectomy with axillary dissection. Histological evaluation revealed grade 3 ductal infiltrating carcinoma (pT2 pN2 M0) with extensive peritumoural vascular invasion. Biological features were: ER-negative and PR-negative; c-erbB2 1+; Ki-67 85%.

Anthracycline-based adjuvant therapy has been shown to be superior to cyclophosphamide–methotrexate–fluorouracil (CMF) regimens in patients with early breast cancer,[10] and the addition of a taxane results in further improvements in survival.[11]

The patient received adjuvant chemotherapy comprising doxorubicin 60 mg/m^2 and cyclophosphamide 600 mg/m^2 every 3 weeks for four cycles, followed by weekly paclitaxel 80 mg/m^2 for 12 administrations until March 2005, with subsequent radiotherapy.

In accordance with the patient, we decided to wait for almost 2 years before considering contralateral prophylactic mastectomy and prophylactic salpingo-oophorectomy, based on increased risk of recurrence during the first years after diagnosis.

In March 2006, she developed pulmonary signs: computed tomography (CT) showed disease progression in the lungs and liver, and involvement of mediastinal lymph nodes.

In patients with metastatic disease, biopsies can be useful to provide histological confirmation of metastasis, characterise biological features of metastatic disease, and guide treatment decisions. In a recent study involving 258 patients, there was discordant receptor expression between primary and recurrent tumours in 13% of cases for ER, 28% for PR, and 5% for human epidermal growth factor receptor-2 (HER2), and biopsy results led to a change of management in 15.9% of patients.[12] In this study, there was no receptor discordance between primary tumour and metastatic lesion in patients with triple-negative disease.

The patient underwent liver biopsy, which revealed that the receptor status of recurrent tissue was the same as in the primary tumour (ER-negative, PR-negative, c-erbB2 1+, Ki-67 85%).

A number of potential treatment strategies for patients with triple-negative metastatic breast cancer are currently available or undergoing evaluation. These include rechallenge with anthracyclines,[13] angiogenesis inhibitors such as bevacizumab,[14] and inhibitors of poly(ADP ribose) polymerase.[15]

The patient received first-line therapy with paclitaxel and bevacizumab.

3. Elderly breast cancer patients

In October 2000, breast cancer was diagnosed following self-palpation in a 70-year-old woman. Bilateral mammography and ultrasound revealed a 2 cm hypoechoic area in the upper internal quadrant of the right breast and a single ipsilateral adenopathy. Nodular core biopsy showed grade 1 lobular infiltrating carcinoma (cT2cN1) that was ER-positive (100%), PR-positive (90%), c-erbB2-negative, and Ki-67 5%. The patient had multiple comorbidities, including hypertension, type 2 diabetes mellitus and severe osteoporosis.

The incidence of breast cancer increases markedly with age: in Europe and the USA, about 50% of new diagnoses are in women aged 65 years and older.[16] However, breast cancer in older patients is associated with more favourable biological features than in younger patients. After adjustment for stage, mortality rates are similar in older and younger women, except for the youngest (<35 years) and oldest (>80 years) age groups, in which survival is lower.[16] Non-breast cancer causes of death are substantial in women aged 70 years or more, even in those with metastases.[17] A comprehensive geriatric assessment (CGA), which includes functional status, comorbidity, socioeconomic issues, polypharmacy, nutritional status, and geriatric syndromes, must be performed before planning treatment, because this can distinguish between fit and unfit patients.[16]

CGA in this case showed that the patient was fit.

In October 2000, the patient underwent right quadrantectomy with axillary dissection, followed by radiotherapy. Histological examination showed grade 2 lobular infiltrating carcinoma (pT2 pN1(2/23)M0) without extensive peritumoural vascular invasion. The tumour was ER-positive (100%), PR-positive (100%) and c-erbB2-negative; Ki-67 expression was 5%.

Both endocrine therapy and systemic chemotherapy may be appropriate for adjuvant treatment in elderly

breast cancer patients, according to tumour biology. Endocrine therapy with tamoxifen produces a 15-year gain of approximately 12% for recurrence-free survival and 9% for breast cancer mortality survival,[10] and a recent meta-analysis has shown that aromatase inhibitors produce significantly lower recurrence rates than tamoxifen, either as initial monotherapy (up-front strategy) or after 2–3 years of tamoxifen (switch strategy).[18] The use of adjuvant chemotherapy more than doubled during the 1990s, with a significant shift towards anthracycline use.[19] Although such treatment has little effect on survival in women with node-negative and hormone receptor-positive tumours, it reduces breast cancer mortality in women with node-positive, hormone receptor-negative tumours.[19]

A recent meta-analysis has examined the impact of non-taxane polychemotherapy in women with ER-poor breast cancer.[20] In women below 69 years of age, chemotherapy significantly reduces the 10-year risk of recurrence and death, with a good safety profile. Furthermore, all patients who received more chemotherapy, irrespective of age, had longer disease-free and overall survival than those who did not. Such findings suggest that age should not be a contraindication to adjuvant chemotherapy in older breast cancer patients who are otherwise in good general health.[21]

While anthracyclines form a mainstay of adjuvant therapy in breast cancer, a number of non-anthracycline regimens have been evaluated. These include treatment with capecitabine[22] and docetaxel plus cyclophosphamide.[23]

This patient's tumour was characterised by high levels of ER, histological grade 2, low proliferation and less than three involved nodes. Moreover, she was a 70-year-old woman with important comorbidities that reduced her life expectancy. It was decided that there were no firm indications for the usefulness of chemotherapy. The decision not to provide chemotherapy in case of luminal A breast cancer at the same stage of the disease is not necessarily consensual, in particular in young women.[24] *The patient received adjuvant therapy with tamoxifen, 20 mg/day for 5 years. In May 2007, after a disease-free interval of about 6.5 years, she developed lung and bone metastases. She was not frail (performance status 1 according to the ECOG score), and had asymptomatic lesions, not at risk for fracture. Her left ventricular ejection fraction (LVEF) was 50%.*

Treatment options for metastatic breast cancer can include endocrine therapy or chemotherapy (anthracycline-based, taxane-based or non-anthracycline-based regimens) alone or associated with bisphosphonates or denosumab (in case of bone involvement).

Aromatase inhibitors form a mainstay of endocrine therapy in patients with metastatic disease. The use of third-generation aromatase inhibitors in the metastatic setting has been shown to produce a significant improvement in overall survival, compared with other endocrine agents.[25] In previously treated patients, the selective ER antagonist fulvestrant has been shown to be comparable in efficacy to aromatase inhibitors.[26,27]

Elderly patients with metastatic breast cancer are expected to derive similar benefits from chemotherapy compared with their younger counterparts in terms of response and delay of progression but, as the risk of toxicity is increased, their quality of life may be impaired during chemotherapy administration.[28] Data from clinical trials comparing monotherapy and polychemotherapy in elderly patients are limited by differences in patient population and study design. However, monotherapy can be considered preferable because of the higher risk of neutropenia in elderly patients, and the high prevalence of comorbidities such as cardiomyopathy, diabetes and renal impairment.[29] A number of strategies can be used to reduce the risk of toxicity in elderly patients, including the use of low-dose cyclophosphamide and methotrexate,[30] oral agents such as capecitabine,[31] or liposomal doxorubicin formulations. Two liposomal doxorubicin formulations are currently available: non-pegylated and pegylated. In clinical trials in patients with metastatic breast cancer, non-pegylated liposomal doxorubicin was comparable in efficacy to conventional doxorubicin, but was associated with significantly less cardiotoxicity.[32–34]

Treatment with bisphosphonates is recommended in elderly patients in case of bone involvement. Such treatment can be offered to frail patients, provided that renal function and dental health are closely monitored.[35] In addition, denosumab has recently been approved by the US Food and Drug Administration for use in this setting (see Perez[36] in this supplement).

Considering visceral involvement, we prescribed chemotherapy with liposomal doxorubicin and cyclophosphamide, in association with a bisphosphonate. In the absence of symptoms, the long disease-free interval and biological features of the disease, another option could be endocrine treatment with an aromatase inhibitor such as letrozole, in association with bisphosphonates. At further relapse without diffuse visceral involvement, a second line of hormonal treatment could be prescribed, fulvestrant being probably the best option.
We would propose chemotherapy only at the onset of clear endocrine resistance of the disease following failure of two subsequent lines of hormonal treatment.

4. HER2-positive breast cancer

In October 2005, a 45-year-old premenopausal woman was diagnosed with right breast cancer following routine mammography. A 1 cm hypoechoic area was present in the right lower quadrant and a 5 mm non-echoic nodule was seen in the right upper external quadrant. No axillary nodes were detectable. A bilateral magnetic resonance of the breast

confirmed two nodules in opposite quadrants with imaging strongly suggestive of cancer. The patient had no family history of breast or ovarian cancer. She was premenopausal with controlled hypertension. Nodule core biopsy of the largest nodule showed grade 2 ductal infiltrating carcinoma (cT1 cN0), and ER, PR and Ki-67 expression were 75%, 60%, and 30%, respectively. c-erbB2 status was 3+.

Trastuzumab with chemotherapy represents the standard of care for women with HER2-positive breast cancer. Following the introduction of this agent, patients with HER2-positive metastatic breast cancer now have improved prognosis compared with women with HER2-negative disease,[37] and it is anticipated that adjuvant therapy with trastuzumab will prevent recurrence in almost 28,000 patients over a 10-year period in the five major European Union countries.[38] The recent NOAH study has shown that the addition of neoadjuvant and adjuvant trastuzumab to neoadjuvant chemotherapy should be considered for women with HER2-positive locally advanced or inflammatory breast cancer to improve event-free survival, survival and clinical and pathological tumour responses. Such a strategy is associated with a low incidence of cardiotoxicity.[39] Furthermore, the NeoSphere[40] and NeoALTTO[41] trials have recently shown superior antitumour activity of anti-HER2 treatment combinations in the neoadjuvant setting.

A right mastectomy with sentinel lymph node biopsy was performed, without prior neoadjuvant therapy, following diagnosis of the bifocal cancer in the right breast. Histological examination of the largest excised nodule confirmed grade 2 ductal infiltrating carcinoma without extensive peritumoural vascular invasion. ER, PR and Ki-67 expression were 70%, 50% and 30%, respectively, c-erbB2 status was 3+. Stage was pT1b(m) pN0 M0. The size of the largest nodule was 8 mm.

Treatment options in patients with HER2-positive early breast cancer must include trastuzumab. Several studies have shown that the addition of trastuzumab significantly improves disease-free survival and overall survival in patients receiving adjuvant chemotherapy.[42–44]

The clinical relevance of HER2 over-expression or amplification was recently studied in a series of 150 consecutive patients with node-negative T1a–b breast cancer.[45] Overall, the 5-year risk of recurrence in these patients was low. However, in women with hormone receptor-positive disease, HER2 over-expression was associated with worse disease-free survival, compared with HER2-negative patients. A further study has reported that women with HER2-positive T1a–b N0 M0 breast cancer are at significant risk of recurrence, and should be considered for adjuvant treatment with anti-HER2 therapies.[46]

The patient received 1 year of adjuvant chemotherapy, consisting of four cycles of doxorubicin and cyclophosphamide, followed by weekly paclitaxel for 12 administrations, and subsequently trastuzumab every 3 weeks for 18 administrations. In May 2006, she started tamoxifen therapy (she was no longer menstruating by this time). In January 2008 (disease-free interval of 2.3 years), mediastinal node involvement and a lung lesion were detected. The patient's performance status was 0, and her LVEF was 60%.

In patients who have previously responded to adjuvant therapy, rechallenge with an agent with proven efficacy is a feasible treatment option. This strategy has been shown to be effective in a number of cancers, including those of the breast, ovary and colon. However, the efficacy of rechallenge treatment is dependent on the duration of the treatment-free interval, and is often limited by cumulative toxicities. A number of retrospective studies have investigated the efficacy of rechallenge with anthracyclines in patients who have previously received adjuvant chemotherapy.[13] These studies showed that previous adjuvant therapy, irrespective of the agent used, had no significant effect on overall or progression-free survival after rechallenge with anthracyclines, suggesting that rechallenge with anthracycline-based regimens is a feasible strategy even in patients who have previously received adjuvant therapy, including anthracyclines.[13]

More recently, a number of phase I/II studies have investigated the use of combinations of liposomal doxorubicin, taxanes and trastuzumab as first-line therapy in HER2-positive patients with metastatic or locally advanced breast cancer who had previously received adjuvant treatment.[47–50] These studies suggest that the combination of non-pegylated liposomal doxorubicin, taxanes and trastuzumab offers promising efficacy in metastatic or locally advanced breast cancer, with acceptable cardiac and general toxicity.

We prescribed three cycles of non-pegylated liposomal doxorubicin, docetaxel and trastuzumab for this patient. Although not approved, this regimen was used following the same regimen delivered in a phase I/II trial carried out at Istituto Scientifico Romagnolo per lo Studio e la Cura dei Tumori (I.R.S.T.) in Meldola, Italy.[48] Another option with approved standard regimens, taking into consideration the long disease-free interval, could be retreatment with weekly paclitaxel[51,52] or paclitaxel and carboplatin, in association with trastuzumab.[53]

5. Conclusions

Breast cancer includes different biological and molecular subtypes requiring different and combined therapeutic approaches. Increasing understanding of the molecular biology of the disease allows us to identify new targeted therapies, which will have important roles in the treatment of specific patient subgroups. Notwithstanding this trend, however, anthracyclines continue to occupy

a central place in the management of breast cancer because of their proven efficacy. The development of liposomal formulations offers a means of maintaining this efficacy while decreasing the cardiotoxicity that is a major limitation to conventional anthracycline therapy.

6. Conflict of interest statement

Dr Gennari and Professor Amadori have both received honoraria from Cephalon. Dr Pietri has no conflicts of interest.

References

1. Morgan D, Sylvester H, Lucas FL, Miesfeldt S. Cancer prevention and screening practices among women at risk for hereditary breast and ovarian cancer after genetic counseling in the community setting. *Fam Cancer* 2009;**8**:277–87.
2. Dent R, Trudeau M, Pritchard KI, et al. Triple-negative breast cancer: clinical features and patterns of recurrence. *Clin Cancer Res* 2007;**13**:4429–34.
3. Dent R, Hanna WM, Trudeau M, Rawlinson E, Sun P, Narod SA. Pattern of metastatic spread in triple-negative breast cancer. *Breast Cancer Res Treat* 2009;**115**:423–8.
4. Cancello G, Maisonneuve P, Rotmensz N, et al. Prognosis and adjuvant treatment effects in selected breast cancer subtypes of very young women (<35 years) with operable breast cancer. *Ann Oncol* 2010;**21**:1974–81.
5. Liedtke C, Mazouni C, Hess KR, et al. Response to neoadjuvant therapy and long-term survival in patients with triple-negative breast cancer. *J Clin Oncol* 2008;**26**:1275–81.
6. Montagna E, Bagnardi V, Rotmensz N, et al. Pathological complete response after preoperative systemic therapy and outcome: relevance of clinical and biologic baseline features. *Breast Cancer Res Treat* 2010;**124**:689–99.
7. Silver DP, Richardson AL, Eklund AC, et al. Efficacy of neoadjuvant cisplatin in triple-negative breast cancer. *J Clin Oncol* 2010;**28**:1145–53.
8. Ryan PD, Tung NM, Isakoff SJ, et al. Neoadjuvant cisplatin and bevacizumab in triple negative breast cancer (TNBC): safety and efficacy. *J Clin Oncol* 2009;**27**(Suppl 15S):abs 551.
9. Torrisi R, Balduzzi A, Ghisini R, et al. Tailored preoperative treatment of locally advanced triple negative (hormone receptor negative and HER2 negative) breast cancer with epirubicin, cisplatin, and infusional fluorouracil followed by weekly paclitaxel. *Cancer Chemother Pharmacol* 2008;**62**:667–72.
10. Early Breast Cancer Trialists' Collaborative Group (EBCTCG). Effects of chemotherapy and hormonal therapy for early breast cancer on recurrence and 15-year survival: an overview of the randomised trials. *Lancet* 2005;**365**:1687–717.
11. Peto R; for the Early Breast Cancer Trialists' Collaborative Group. The worldwide overview: new results for systemic adjuvant therapies. 30th Annual San Antonio Breast Cancer Symposium, San Antonio, Texas, USA, 13–16 December 2007:plenary lecture 1.
12. Amir E, Clemons M, Freedman OC, et al. Tissue confirmation of disease recurrence in patients with breast cancer: pooled analysis of two large prospective studies. *J Clin Oncol* 2010;**28**(Suppl 15s):abs 1007.
13. Morabito A, Piccirillo MC, Monaco K, et al. First-line chemotherapy for HER-2 negative metastatic breast cancer patients who received anthracyclines as adjuvant treatment. *Oncologist* 2007;**12**:1288–98.
14. Miles D, Chan A, Dirix LY, et al. Phase III study of bevacizumab plus docetaxel compared with placebo plus docetaxel for the first-line treatment of human epidermal growth factor receptor 2-negative metastatic breast cancer. *J Clin Oncol* 2010;**28**:3239–47.
15. O'Shaughnessy J, Osborne C, Pippen J, et al. Efficacy of BSI-201, a poly(ADP-ribose) polymerase-1 (PARP1) inhibitor, in combination with gemcitabine/carboplatin (G/C) in patients with metastatic triple-negative breast cancer (TNBC): results of a randomized phase II trial. *J Clin Oncol* 2009;**27**(Suppl 18s):abs 3.
16. Muss HB. Adjuvant treatment of elderly breast cancer patients. *Breast* 2007;**16**(Suppl 2):S159–65.
17. Yancik R, Wesley MN, Ries LA, Havlik RJ, Edwards BK, Yates JW. Effect of age and comorbidity in postmenopausal breast cancer patients aged 55 years and older. *JAMA* 2001;**285**:885–92.
18. Dowsett M, Cuzick J, Ingle J, et al. Meta-analysis of breast cancer outcomes in adjuvant trials of aromatase inhibitors versus tamoxifen. *J Clin Oncol* 2010;**28**:509–18.
19. Giordano SH, Duan Z, Kuo YF, Hortobagyi GN, Goodwin JS. Use and outcomes of adjuvant chemotherapy in older women with breast cancer. *J Clin Oncol* 2006;**24**:2750–6.
20. Early Breast Cancer Trialists' Collaborative Group (EBCTCG). Adjuvant chemotherapy in oestrogen-receptor-poor breast cancer: patient-level meta-analysis of randomised trials. *Lancet* 2008;**371**:29–40.
21. Muss HB, Woolf S, Berry D, et al. Adjuvant chemotherapy in older and younger women with lymph node-positive breast cancer. *JAMA* 2005;**293**:1073–81.
22. Muss HB, Berry DA, Cirrincione CT, et al. Adjuvant chemotherapy in older women with early-stage breast cancer. *N Engl J Med* 2009;**360**:2055–65.
23. Jones S, Holmes FA, O'Shaughnessy J, et al. Docetaxel with cyclophosphamide is associated with an overall survival benefit compared with doxorubicin and cyclophosphamide: 7-year follow-up of US Oncology Research Trial 9735. *J Clin Oncol* 2009;**27**:1177–83.
24. Goldhirsch A, Wood WC, Coates AS, Gelber RD, Thürlimann B, Senn HJ; Panel members. Strategies for subtypes – dealing with the diversity of breast cancer: highlights of the St Gallen International Expert Consensus on the Primary Therapy of Early Breast Cancer 2011. *Ann Oncol* 2011;**22**:1736–47.
25. Biganzoli L, Licitra S, Claudino W, Pestrin M, Leo AD. Clinical decision making in breast cancer: TAM and aromatase inhibitors for older patients – a jungle? *Eur J Cancer* 2007;**43**:2270–8.
26. Chia S, Gradishar W, Mauriac L, et al. Double-blind, randomized placebo controlled trial of fulvestrant compared with exemestane after prior nonsteroidal aromatase inhibitor therapy in postmenopausal women with hormone receptor-positive, advanced

breast cancer: results from EFECT. *J Clin Oncol* 2008;**26**:1664–70.
27. Howell A, Robertson JF, Quaresma Albano J, et al. Fulvestrant, formerly ICI 182,780, is as effective as anastrozole in postmenopausal women with advanced breast cancer progressing after prior endocrine treatment. *J Clin Oncol* 2002;**20**:3396–403.
28. Wildiers H, Kunkler I, Biganzoli L, et al. Management of breast cancer in elderly individuals: recommendations of the International Society of Geriatric Oncology. *Lancet Oncol* 2007;**8**:1101–15.
29. Aapro M, Monfardini S, Jirillo A, Basso U. Management of primary and advanced breast cancer in older unfit patients (medical treatment). *Cancer Treat Rev* 2009;**35**: 503–8.
30. Colleoni M, Rocca A, Sandri MT, et al. Low-dose oral methotrexate and cyclophosphamide in metastatic breast cancer: antitumor activity and correlation with vascular endothelial growth factor levels. *Ann Oncol* 2002;**13**:73–80.
31. Blum JL, Jones SE, Buzdar AU, et al. Multicenter phase II study of capecitabine in paclitaxel-refractory metastatic breast cancer. *J Clin Oncol* 1999;**17**:485–93.
32. Batist G, Ramakrishnan G, Rao CS, et al. Reduced cardiotoxicity and preserved antitumor efficacy of liposome-encapsulated doxorubicin and cyclophosphamide compared with conventional doxorubicin and cyclophosphamide in a randomized, multicenter trial of metastatic breast cancer. *J Clin Oncol* 2001;**19**:1444–54.
33. Batist G, Harris L, Azarnia N, Lee LW, Daza-Ramirez P. Improved anti-tumor response rate with decreased cardiotoxicity of non-pegylated liposomal doxorubicin compared with conventional doxorubicin in first-line treatment of metastatic breast cancer in patients who had received prior adjuvant doxorubicin: results of a retrospective analysis. *Anticancer Drugs* 2006;**17**:587–95.
34. Harris L, Batist G, Belt R, et al. Liposome-encapsulated doxorubicin compared with conventional doxorubicin in a randomized multicenter trial as first-line therapy of metastatic breast carcinoma. *Cancer* 2002;**94**:25–36.
35. Body JJ, Coleman R, Clezardin P, et al. International Society of Geriatric Oncology (SIOG) clinical practice recommendations for the use of bisphosphonates in elderly patients. *Eur J Cancer* 2007;**43**:852–8.
36. Perez EA. New treatment strategies in the management of breast cancer. *EJC Suppl* 2011;**9**(2):22–9.
37. Dawood S, Broglio K, Buzdar AU, Hortobagyi GN, Giordano SH. Prognosis of women with metastatic breast cancer by HER2 status and trastuzumab treatment: an institutional-based review. *J Clin Oncol* 2010;**28**:92–8.
38. Weisgerber-Kriegl U, Cirrincione A, McNiven P. Estimation of the epidemiological effect of trastuzumab over 10 years in 5 European countries. *J Clin Oncol* 2008;**26**(Suppl):abs 6589.
39. Gianni L, Eiermann W, Semiglazov V, et al. Neoadjuvant chemotherapy with trastuzumab followed by adjuvant trastuzumab versus neoadjuvant chemotherapy alone, in patients with HER2-positive locally advanced breast cancer (the NOAH trial): a randomised controlled superiority trial with a parallel HER2-negative cohort. *Lancet* 2010;**375**:377–84.
40. Gianni L, Pienkowski T, Im Y-H, et al. Neoadjuvant pertuzumab (P) and trastuzumab (H): antitumor and safety analysis of a randomized phase II study ('NeoSphere'). 33rd Annual San Antonio Breast Cancer Symposium, San Antonio, Texas, USA, 8–12 December 2010:abs S3-2.
41. Baselga J, Bradbury I, Eidtmann H, et al. First results of the NeoALTTO trial (BIG 01-06/EGF 106903): a phase III, randomized, open label, neoadjuvant study of lapatinib, trastuzumab, and their combination plus paclitaxel in women with HER2-positive primary breast cancer. 33rd Annual San Antonio Breast Cancer Symposium, San Antonio, Texas, USA, 8–12 December 2010:abs S3-3.
42. Smith I, Procter M, Gelber RD, et al. 2-year follow-up of trastuzumab after adjuvant chemotherapy in HER2-positive breast cancer: a randomised controlled trial. *Lancet* 2007;**369**:29–36.
43. Slamon D, Eiermann W, Robert N, et al. Phase III randomized trial comparing doxorubicin and cyclophosphamide followed by docetaxel (AC → T) with doxorubicin and cyclophosphamide followed by docetaxel and trastuzumab (AC → TH) with docetaxel, carboplatin and trastuzumab (TCH) in Her2neu positive early breast cancer patients: BCIRG 006 study. *Cancer Res* 2009;**69**(24 Suppl):abs 62.
44. Perez E, Suman V, Davidson N, et al. Results of chemotherapy alone, with sequential or concurrent addition of 52 weeks of trastuzumab in the NCCTG N9831 HER2-Positive Adjuvant Breast Cancer Trial. *Cancer Res* 2009;**69**(24 Suppl):abs 80.
45. Curigliano G, Viale G, Bagnardi V, et al. Clinical relevance of HER2 overexpression/amplification in patients with small tumor size and node-negative breast cancer. *J Clin Oncol* 2009;**27**:5693–9.
46. Gonzalez-Angulo AM, Litton JK, Broglio KR, et al. High risk of recurrence for patients with breast cancer who have human epidermal growth factor receptor 2-positive, node-negative tumors 1 cm or smaller. *J Clin Oncol* 2009;**27**:5700–6.
47. Cortes J, Di Cosimo S, Climent MA, et al. Nonpegylated liposomal doxorubicin (TLC-D99), paclitaxel, and trastuzumab in HER-2-overexpressing breast cancer: a multicenter phase I/II study. *Clin Cancer Res* 2009;**15**:307–14.
48. Amadori D, Milandri C, Comella G, et al. A phase I/II trial of non-pegylated liposomal doxorubicin, docetaxel and trastuzumab as first-line treatment in HER-2-positive locally advanced or metastatic breast cancer. *Eur J Cancer* 2011 Jun 10 [Epub ahead of print]. doi: 10.1016/j.ejca.2011.05.005.
49. Venturini M, Bighin C, Puglisi F, et al. A multicentre Phase II study of non-pegylated liposomal doxorubicin in combination with trastuzumab and docetaxel as first-line therapy in metastatic breast cancer. *Breast* 2010;**19**:333–8.
50. Anton A, Ruiz A, Plazaola A, et al. Phase II clinical trial of liposomal-encapsulated doxorubicin citrate and docetaxel, associated with trastuzumab, as neoadjuvant treatment in stages II and IIIA HER2-overexpressing breast cancer patients. GEICAM 2003-03 study. *Ann Oncol* 2010 Jul 5 [Epub ahead of print]. doi: 10.1093/annonc/mdq317.
51. Seidman AD, Berry D, Cirrincione C, et al. Randomized phase III trial of weekly compared with every-3-weeks paclitaxel for metastatic breast cancer, with trastuzumab for all HER-2 overexpressors and random assignment to trastuzumab or not in HER-2 nonoverexpressors: final results of Cancer and Leukemia Group B protocol 9840. *J Clin Oncol* 2008;**26**:1642–9.

52. Gori S, Colozza M, Mosconi AM, et al. Phase II study of weekly paclitaxel and trastuzumab in anthracycline- and taxane-pretreated patients with HER2-overexpressing metastatic breast cancer. *Br J Cancer* 2004;**90**:36–40.

53. Robert N, Leyland-Jones B, Asmar L, et al. Randomized phase III study of trastuzumab, paclitaxel, and carboplatin compared with trastuzumab and paclitaxel in women with HER-2-overexpressing metastatic breast cancer. *J Clin Oncol* 2006;**24**:2786–92.