Journal of BUON **17:** 209-222, 2012

REVIEW ARTICLE

Promising therapeutic options in triple-negative breast cancer

A. Bilici[1], C. Arslan[2], K. Altundag[2]

[1]Sisli Etfal Research and Training Hospital, Department of Medical Oncology, Istanbul; [2]Department of Medical Oncology, Hacettepe University Institute of Oncology, Ankara, Turkey

Summary

Triple-negative breast cancer (TNBC) has a greater risk of recurrence despite more aggressive therapy even in low-risk category. TNBC is high grade, hormone receptor and HER-2 negative, it exhibits a high level of Ki-67 staining and expresses the epithelial growth factor receptor (EGFR). Because of its expression profile, treatment options are limited to cytotoxic chemotherapy. Molecular defects that give rise to BRCA1-associated breast cancer also occur in TNBC. Thus, the combination of poly-(ADP-ribose)-polymerase (PARP) inhibitors with drugs that cause DNA breakages, such as alkylating agents and topoisomerase I inhibitors, could theoretically potentiate the efficacy of each drug in patients with TNBC. Clinical trials with various targeted approaches alone or in combination with different chemotherapeutic agents are currently underway. In this review, current and future treatment approaches in TNBC with novel targeted agents are discussed.

Key words: basal-like, breast cancer, treatment, triple negative

Introduction

TNBC is characterized by lack of estrogen receptor (ER) and progesterone receptor (PR) expression, as well as absence of human epidermal growth factor receptor-2 (HER-2) overexpression upon immunohistochemical analysis [1]. Despite the fact that the terms TNBC and basal-like cancer are often used interchangeably, there is a substantial overlap in the biological and clinical characteristics of basal-like breast cancer and TNBC; thus, they are different subtypes of cancer [1,2]. Basal-like breast cancer constitutes one of the 5 subtypes of breast cancer and is diagnosed using microarray-based expression analysis [2,3]. This subtype comprises a heterogeneous group of tumors defined by absence or low level of expression of ER and PR and a very low prevalence of HER-2 overexpression. In basal-like breast cancer the expression of genes is usually detected in the basal or myoepithelial cells of the human breast [3]. These tumors stain positively for basal cell cytokeratins (CK5, 6, 17) [4].

Approximately 15-20% of breast cancers are TNBCs [5]. TNBCs are characterized by an aggressive clinical course and poor survival, despite more aggressive therapy, even in low-risk category [6]. High rates of TNBC have been observed in young women, which may be associated with a greater likelihood of BRCA1 expression; TNBC is common in premenopausal African or Hispanic women and in lower socioeconomic groups. The histological features of triple-negative tumors include high grade, high Ki-67 index, and necrosis [7-9]. TNBCs frequently metastasize via the hematogeneous route rather than via the lymphatics, and thus shows less axillary lymph node metastasis compared to non-TNBC [9,10]. Patients with TNBC have a greater risk of recurrence and once metastatic TNBC is present a much shorter median time from relapse to death is commonly observed [5]. Patients with germline mutations in BRCA genes are at risk of developing breast, ovarian, pancreatic, and prostate cancers, among other malignancies. The products of BRCA genes have a variety of roles and cells that lack a functional BRCA1 or BRCA2 display deficiency in the repair of DNA double-strand breaks [11]. The rate of BRCA1 mutation is approximately 2% in all women with breast cancer [12], but its frequency can be as high as 10% in TNBC

Correspondence to: Kadri Altundag, MD. Department of Medical Oncology, Hacettepe University Institute of Oncology, Sihhiye Ankara 06100, Turkey. Tel: +90 312 3052954, Fax: +90 324 2009, E-mail: altundag66@yahoo.com

Received 09-09-2011; Accepted 02-10-2011

patients [13]. In addition, TNBC is the major tumor type in BRCA1 mutation carriers [14,15]. Patients with TNBC show many similarities to BRCA1-associated breast cancer patients: both BRCA-related breast cancer and TNBC have aggressive clinical courses, they share pathological and clinical features and they both have a high histological grade, are hormone-receptor and HER-2 negative, they exhibit a high level of Ki-67 staining, they express the epithelial growth factor receptor (EGFR) and CK5/6, and they harbor a p53 mutation [16,17].

TNBC is not suitable for treatment with hormone therapy or the anti-HER-2 monoclonal antibody trastuzumab because of its biological expression profile. Therefore, the treatment options are currently limited to cytotoxic chemotherapy [5,7,18,19]. Chemotherapy remains the mainstay of TNBC treatment. Previous studies associated with neoadjuvant chemotherapy showed that TNBC was more sensitive to chemotherapy than hormone-receptor positive subtypes, as indicated by higher pathological complete response (pCR) rates [20,21]. Despite this chemosensitivity, TNBC is associated with shorter 5-year survival than non-TNBC phenotypes [7]. Therefore, molecular therapeutic targets are promptly required to improve the survival of these patients. In this article, we review the currently available and promising therapeutic options that are still under development for patients with TNBC.

Current treatment strategies for TNBC

Chemotherapy is currently the standard option for the systemic treatment of TNBC due to lack of molecular targets, such as the absence of ER, PR, and HER-2 [5,18,19]. Many chemotherapeutic agents have been used, including anthracyclines, taxanes, ixabepilone, and platinum agents. Several studies indicated that different chemotherapeutic agents were found to be useful against TNBC in the adjuvant setting [22,23]. Moreover, neoadjuvant chemotherapy trials have shown that TNBC has a better response rate and a more frequent rate of pCR [20,24]. However, this does not improve the survival of patients. Selected clinical trials with respect to chemotherapeutic agents in TNBC are summarized in Table 1.

Chemotherapy

Platinum agents

The beneficial effects of cisplatin or carboplatin in the treatment of TNBC are currently being assessed in clinical trials that are partly based on their ability to bind directly to DNA. TNBC shares similar features with BRCA1-associated breast cancer; a dysfunction in BRCA1 and its pathway is associated with a specific DNA-repair defect that sensitizes cells to platinum drugs in animal models [25,26].

In a small retrospective study performed by Bryski et al. [27], neoadjuvant cisplatin treatment showed a higher rate of pCR (83%) than the other regimens, including non-platinum drugs. In a study by Silver et al. [28], neoadjuvant single-agent cisplatin resulted in a 22% pCR in 28 patients with TNBC. Cisplatin has also been used with other cytotoxic agents as combination chemotherapy in the neoadjuvant setting. In their study [29] Torrisi et al. showed 86% overall response and 40% pCR rates obtained with 4 cycles of neoadjuvant epirubicin, cisplatin, and 5-fluorouracil chemotherapy, followed by 3 cycles of weekly paclitaxel. Frasci et al. evaluated neoadjuvant dose-dense cisplatin, epirubicin and paclitaxel with G-CSF support in 74 patients with operable, large TNBC. The pCR rate was remarkably high (62%), while the 3-year disease-free survival (DFS) rate exceeded 80% [30]. In addition, this regimen increased the rate of breast-conserving surgery. These are encouraging results that merit further validation and testing. The role of platinum agents in the neoadjuvant setting for TNBC patients is currently being tested by 2 randomized studies (CALGB 40603 and the Spanish Breast Cancer Research Group study). In both of these trials, patients will be randomized to receive carboplatin as a part of neoadjuvant chemotherapy; in the Spanish study they will be treated with 4 courses of epirubicin and cyclophosphamide and thereafter will be randomized to receive docetaxel or carboplatin (NCT00432172).

Cisplatin has been found to be effective in patients with metastatic disease. In one study, platinum-containing chemotherapy as first- or second-line treatment was investigated in 106 metastatic breast cancer patients. The rate of TNBC was 36 (34%) and platinum-based chemotherapy resulted in a 39% overall response rate (ORR) and a 67% disease control (DC) rate. Although the ORR and DC rates were similar to those obtained for other phenotypes, the overall survival (OS) was shorter [31]. In another study, Sirohi et al. retrospectively analyzed 94 (17 with TNBC), 79 (11 with TNBC) and 155 (34 with TNBC) patients who were treated with platinum-containing chemotherapy in neoadjuvant, adjuvant and metastatic settings, respectively [32]. The treatment regimens included epirubicin; cisplatin/carboplatin and 5-fluorouracil; or mitomycin-C, vinblastine, and cisplatin/carboplatin. The authors reported

Table 1. Selected clinical trials of chemotherapeutic agents in patients with triple-negative breast cancer

	Setting	*Regimen*	*Study population*	*Results/Comments*
Platinum agents				
Bryski et al. [27]	Neoadjuvant	Cisplatin	BRCA-1 positive breast cancer	pCR rate was higher (83%) than other regimens, including non-platinum drugs.
Silver et al. [28]	Neoadjuvant	Cisplatin	Stage II and III TNBC	pCR rate 22%.
Torrisi et al. [29]	Neoadjuvant	ECFx4→Px3 / neoadjuvant+adj. CM P.O.	T2-3, N0-3; TNBC	ORR 86%, pCR rate 40%, 2-year DFS rate 87.5%.
Frasci et al. [30]	Neoadjuvant	Weekly cisplatin/E/P+G-CSFx8 weeks	Large operable TNBC	The pCR rate was remarkably high (62%), while the 3-year DFS rate was 80%. Increased rate of BCS (67.5%)
Yi et al. [31]	Metastatic	Platinum-based chemotherapy	Metastatic breast cancer (TNBC n: 36)	ORR 39% and disease control rate 67%. OS was shorter in TNBC than non-TNBC.
Sirohi et al. [32]	Neoadjuvant, adjuvant and metastatic	Platinum-containing chemotherapy	Early and metastatic breast cancer	CR for TNBC was increased in neoadjuvant setting (88 vs 51%), but 5-year OS rate for TNBC in 3 settings was worse than non-TNBC (64%, 44% and 79%). Five-year DFS was shorter in TNBC patients compared with non-TNBC (57 vs 72%).
Uhm et al. [33]	Metastatic/ first- or second-line	Platinum+paclitaxel	Metastatic breast cancer	ORR rate 39% for TNBC. OS in TNBC was shorter than non-TNBC (21 vs 56 months).
Isakoff et al. [34]	Metastatic	Platinum	TNBC	ORR rate 30.2% and median PFS interval 89 days.
Alkylating agents and taxanes				
Liedtke et al. [20]	Neoadjuvant	FAC/FEC/ AC±taxane	Locally advanced breast cancer	pCR rate was significantly higher for TNBC than non-TNBC (22 vs 11%). 3-year OS and PFS rates 74 vs 89% and 63 vs 76%, respectively in TNBC vs non-TNBC.
Carey et al. [21]	Neoadjuvant	AC	Locally advanced breast cancer	pCR was 27 vs 36 vs 7% in basal-like vs Her(+)/ ER(-) vs luminal breast cancer, respectively.
Torrisi et al. [29]	Neoadjuvant	ECF x4→Px3/ neoadjuvant+adj. po CM	T2-3, N0-3; TNBC	ORR 86%, pCR rate 40%, 2-year DFS rate 87.5%.
Hugh et al. [36]	Adjuvant	TAC vs FAC	Early breast cancer	There was marginal significance in TNBC cohort, with 3-year DFS 73.5 vs 60% for TAC and FAC, respectively.
Rouzier et al. [24]	Neoadjuvant	Weekly paclitaxel x12 weeks→FACx4	Locally advanced breast cancer	pCR rate 45 vs 45 vs 6% in basal-like vs Her-2(+) vs luminal breast cancer.
Sanchez-Munoz et al. [39]	Neoadjuvant	Arm A: Ecx3→GP±T Arm B: APG±T	Locally advanced breast cancer	pCR rate in TNBC was higher than Her-2(+)/HR(-) and Her-2(-)/HR(+) cohorts (58.3%, 32% and 5.4%, respectively), but DFS rates were similar.
Bidard et al. [43]	Neoadjuvant	FEC/FAC	Breast cancer treated with neoadjuvant chemotherapy	pCR rate 17 vs 4% in TNBC vs non-TNBC. p53 immunostaining was associated with a trend for a higher rate of pCR in TNBC.
Anti-tubulin agents				
Thomas et al. [47]	Metastatic	Arm A: Ixabepilone+cap Arm B: cap	Breast cancer failed to respond to anthracyclines + taxanes	Arm A showed higher RR (27 vs 9%) and longer TTP (4.1 vs 2.1 months) in TNBC subgroup.
Roche et al. [48]	Metastatic	Ixabepilone+cap vs cap	Breast cancer pretreated anthracyclines + or taxanes	ORR 31 vs 15%, PFS rate 4.2 vs 1.7 months in ixabepilone+cap vs cap monotherapy for TNBC cohort.

A: adriamycin, BCS: breast-conserving surgery, C: cyclophosphamide, Cap: capecitabine, T: docetaxel, DFS: disease-free survival, E: epirubicin, ECF: epirubicin, cisplatin, and 5-fluorouracil, ER: estrogen receptor, F: 5-fluorouracil, G: gemcitabine, G-CSF: granulocyte-colony-stimulating factor, HR: hormone receptor, M: methotrexate, ORR: overall response rate, OS: overall survival, P: paclitaxel, pCR: pathological complete response, PFS: progression-free survival, RR: response rate, TTP: time to progression, TNBC: triple-negative breast cancer

that the complete response (CR) rate for TNBC was increased with platinum-based chemotherapy compared to others in the neoadjuvant setting (88 vs. 51%), but they reported a worse 5-year OS rate following neoadjuvant and adjuvant chemotherapy for TNBC patients than patients with non-TNBC (64, 44, and 79%, respectively) in early breast cancer. Moreover, the 5-year DFS rate for TNBC was shorter than that of patients with non-TNBC (57 vs. 72%). In a phase II study carried out by Uhm et al., the ORR rate was found to be 37.5% with a platinum and paclitaxel combination as first- or second-line treatment in 36 metastatic TNBC patients [33]. Despite these trials demonstrating the activity of platinum-based chemotherapy in the metastatic setting, their major limitation was the small sample size.

There are a number of studies for early, locally-advanced or metastatic TNBC patients currently under investigation or planned for platinum compounds combined with taxanes, gemcitabine and/or bevacizumab, sunitinib, and new agents (NCT00532727, NCT00887575, NCT01150513, NCT01238133, NCT00691379, NCT01207102). In addition, the results of the phase II Translational Breast Cancer Research Consortium 009 trial presented at the 2011 ASCO annual meeting showed that the ORR was 30.2% and the median progression free survival (PFS) time was 89 days with single-agent platinum in metastatic TNBC [34]. However, the expression of p63/p73 as a biomarker of platinum sensitivity is ongoing. In addition, a phase III study is currently evaluating carboplatin or docetaxel with a crossover upon progression in 400 TNBC patients (NCT00532727). The current trials will help clarify the role of platinum agents and the relationship between platinum sensitivity and BRCA1 mutations.

Alkylating agents and taxanes

In patients with TNBC, other cytotoxic regimens have also been found to be active, showing that TNBC is a chemosensitive tumor. Despite its chemosensitivity, TNBC is still related to a poor prognosis. In a prospective study from the M.D. Anderson Cancer Center, neoadjuvant anthracycline-based combinations with or without taxanes yielded a pCR rate of 22% in 57 out of 255 patients with TNBC, which was significantly higher than the 11% rate seen among patients with non-TNBC phenotypes [35]. However, the 3-year OS was significantly shorter compared with patients without TNBC. High clinical response rates with anthracycline-based regimens were also obtained in other studies [21,27]. In the National Surgical Adjuvant Breast and Bowel Project (NSABP) trial, the doxorubicin-cyclophosphamide-taxane combination in the preoperative setting was found to be effective with high in-breast response rates for the TNBC group [21]. In another larger, retrospective taxane-5-fluorouracil-doxorubicin series, the outcomes of patients with TNBC were compared to those of non-TNBC patients [29]. This study showed that although patients with TNBC had a higher pCR rate than non-TNBC patients (22 vs. 11%), they had significantly shorter 3-year PFS and OS rates (63 vs. 76% and 74 vs. 89%, respectively). In a meta-analysis performed by Di Leo et al., as well as in smaller phase II and III studies with anthracyclines, different results were reported by individual agents and regimens for patients with TNBC [35].

The effect of adjuvant anthracyclines plus taxane in TNBC was investigated in other studies. The Breast Cancer International Research Group (BCIRG) 001 trial compared docetaxel-doxorubicin-cyclophosphamide (TAC) vs. 5-fluorouracil-doxorubicin-cyclophosphamide (FAC) [36]. This study showed that the addition of taxane (TAC) resulted in an advantageous high response rate in the triple-negative cohort. In addition, anthracycline and taxane combinations were found to be useful, with high pCR rates in the neoadjuvant setting for patients with breast cancer [37, 38], but there is no head-to-head comparison study of neoadjuvant chemotherapy with and without taxanes in patients with TNBC. However, some retrospective series reported that the addition of taxanes to anthracyclines yielded high pCR rates in TNBC patients [20, 24, 39]. In their study Rouzier et al. indicated that weekly paclitaxel for 12 weeks followed by FAC for 4 cycles increased the pCR rate to 45%. In this study, patients with basal-like and HER-2-positive breast cancer had the highest pCR rates compared to those with luminal and normal-like breast cancer [24]. Sanchez-Munoz et al. found that anthracycline-plus-taxane-containing chemotherapy was associated with superior response rates for patients with TNBC than for those with HER-2 and hormone receptor (HR)-positive disease in the neoadjuvant setting (pCR rates of 58.3, 32 and 5.4%, respectively). On the other hand, no difference was detected with respect to DFS between these groups [39].

The predictive values of some markers for chemotherapy response are being investigated in TNBC. The topoisomerase-2α (TOPO2A) gene has been found to be molecular target for anthracycline therapy and it is located next to the HER-2 gene on chromosome 17q12-q21. It was found that TOPO2A gene amplification was related to anthracycline sensitivity in breast cancer, but the rate of TOPO2A gene amplification was only detected in 1-10% of patients with TNBC [40,41]. The poor prognosis of TNBC might be related to these low rates of TOPO2A amplification despite anthracycline chemotherapy. Tumor protein 53 (TP53) is a tumor suppressor gene that might contribute to cancer progression be-

cause it is responsible for the cellular response to DNA damage [42]. In their study Bidard et al., who investigated the presence of p53 by immunohistochemistry (IHC) staining in 296 breast cancer patients, showed that p53 was found in 59% of TNBC patients and that it was associated with poor tumor differentiation. Moreover, the authors found that p53-positive tumors responded better to alkylating neoadjuvant chemotherapy than TNBC patients with p53-negative tumors (20 vs. 10%). In addition, the positivity of p53 was correlated with a trend toward a higher rate of pCR in TNBC compared to non-TNBC patients [43].

Anti-tubulin agents

Epothilones bind β-tubulin, stabilize microtubules, and result in cell cycle arrest in the G2/M phase and apoptosis, similar to taxanes. Ixabepilone is a new agent that bypasses the resistance mechanism associated with drug efflux pumps and specific paclitaxel resistance related to β-tubulin [44]. It has recently been approved for the treatment of taxane-refractory metastatic breast cancer, and in combination with capecitabine for patients with advanced breast cancer refractory to anthracyclines and taxanes. Ixabepilone has been evaluated in subsets of patients with metastatic TNBC in phase II and phase III trials [45,46]. A study on the use of ixabepilone plus capecitabine vs. capecitabine monotherapy in patients who failed to respond to anthracyclines plus taxanes showed a higher response rate (27 vs. 9%) and a longer time to progression (4.1 vs. 2.1 months) for the combination in the triple-negative subgroup [47]. Furthermore, the pooled results of the 046 study (taxane resistant) and the 048 study (population pretreated with anthracyclines and taxanes) were presented at the 2008 San Antonio Breast Cancer Symposium [48]. The ixabepilone plus capecitabine combination indicated that benefits were found in terms of ORR (31 vs. 15%) and PFS (4.2 vs. 1.7 months), but not for OS (10.3 vs. 9.0 months), in patients with advanced TNBC. The ongoing phase III adjuvant study PACS-08, which stratifies TNBCs, includes the use of combination chemotherapy followed by docetaxel or ixabepilone (NCT00630032).

Targeted therapies

TNBC does not express ER, PR, or HER-2 gene products; therefore, it does not respond to endocrine therapy or other available targeted agents. However, numerous targeted agents are under development for patients with TNBC. Table 2 summarizes selected clinical studies of targeted therapies in patients with TNBC.

Bevacizumab

Bevacizumab, a monoclonal antibody directed against the vascular endothelial growth factor (VEGF), was approved by the US Food and Drug Administration as a first-line treatment in metastatic breast cancer in combination with paclitaxel, as it prolonged the PFS (8.8 vs. 4.6 months) and increased response rates compared to paclitaxel monotherapy (36.9 vs. 21.2%) in the phase III E2100 study, but the OS was similar in both groups. This PFS benefit with the addition of bevacizumab to paclitaxel was maintained in the triple-negative subgroup [49]. Two additional phase III trials indicated an increased ORR with the addition of bevacizumab in metastatic cancer: the phase III study AVADO combined bevacizumab with docetaxel [50], and the RIBBON-1 study used bevacizumab in combination with different drugs (capecitabine, nabpaclitaxel, docetaxel, or anthracyclines) [51]. The subgroup analysis of these 3 phase III studies was presented at the 2009 San Antonio Breast Cancer Symposium [52]. This analysis showed that the addition of bevacizumab to chemotherapy improved the PFS regardless of the chemotherapy backbone used in metastatic TNBC patients. On the other hand, in a study performed by Balduzzi et al., a 33% pCR rate and 54% PR was reported in patients with locally advanced breast cancer with neoadjuvant epirubicin and cisplatin plus infusional fluorouracil for 4 cycles following 3 courses of weekly paclitaxel in combination with bevacizumab [53].

Based on these results, novel protocols are ongoing that include bevacizumab in combination with different adjuvant chemotherapy regimens in only TNBC (NCT00528567 BEATRICE) or HER-2-negative tumors (CALGB 40603 NCT00861705). The BEATRICE trial is a phase III adjuvant trial that recently completed patient recruitment; its results are being awaited.

EGFR inhibitors

The expression of EGFR is more frequent in TNBC than in other subtypes and it may be a viable target in TNBC treatment [4]. Cetuximab is a chimeric monoclonal antibody that inhibits EGFR. It was evaluated both alone and in combination with carboplatin in a randomized phase II trial. The TBCRC 001 trial was a phase II trial involving 102 patients with metastatic basal-like TNBC who had received prior chemotherapy. Although cetuximab monotherapy showed low activity (6% response rate, 4% stable disease for >6 months and 10% clinical benefit), cetuximab plus carboplatin showed higher rates of partial response (18%) and clinical benefit (27%). However, most patients progressed

Table 2. Targeted therapies in triple-negative breast cancer

	Phase/Setting	*Treatment*	*Study population*	*Results/Comments*
Anti-VEGF agents				
Bevacizumab				
Miller et al. [49]	III /Metastatic	Weekly paclitaxel, d 1,8,15 vs bevacizumab 10 mg/kg, d 1 and 15+paclitaxel, q3 weeks	Previously untreated metastatic breast cancer	The addition of bevacizumab increased PFS (8.8 vs 4.6 months), but not OS, as compared with paclitaxel monotherapy in TNBC cohort.
Miles et al. [50]	III / Recurrent or metastatic-first-line	Docetaxel+placebo vs Docetaxel + bevacizumab 7.5 or 15 mg/kg, q3 weeks	Locally recurrent and metastatic breast cancer	PFS was significantly better for bevacizumab 15 mg/kg plus docetaxel than docetaxel plus placebo (8.1 vs 6.0 months). It had similar benefit in both HR(+) and HR (-) subgroups.
Robert et al. [51]	III / Recurrent or metastatic-first-line	Capecitabine 2000 mg/m^2 for 4 days, nab-paclitaxel 260 mg/m^2, docetaxel 75-100 mg/m^2, or doxorubicin or epirubicin q3 weeks± bevacizumab 15 mg/kg or placebo q3w	Locally recurrent and metastatic breast cancer	Median PFS was 6.1 vs 4.2 in capecitabine group and 14.5 vs 8.2 in taxane/anthracycline group with the addition of bevacizumab.
Balduzzi et al. [53]	III/ Neoadjuvant	ECFx4→weekly paclitaxel + bevacizumab x3	Locally advanced breast cancer	pCR 33% and PR 54%.
EGFR inhibitors				
Cetuximab				
Carey et al. [54]	II / Metastatic	Cetuximab ± carboplatin	Metastatic TNBC who had received prior chemotherapy	PR 18% and clinical benefit 27% in cetuximab+ carboplatin group, but most patients progressed rapidly.
O'Shaughnessy et al. [55]	II / Metastatic	Weekly irinotecan + carboplatin ± cetuximab	Metastatic breast cancer	A higher RR was obtained with the addition of cetuximab to chemotherapy (49 vs 30%) in TNBC subgroup.
Baselga et al. [56]	II /Metastatic	Cisplatin ± cetuximab	Metastatic TNBC	ORR was 20 vs 10.3% in cetuximab+cisplatin vs cisplatin alone. PFS was shorter in cisplatin alone compared with cetuximab combination (1.5 vs 3.7) months.
Panitumumab				
Nabholtz et al. (60)	II / Neoadjuvant	Panitumumab + FEC100→docetaxel	Locally advanced breast cancer	pCR was 17% and overall CRR 80%. Conservative surgery was performed in 87% of the patients.
Tyrosine kinase inhibitors				
Sunitinib				
Burstein et al. [64]	II / Metastatic	Sunitinib 50 mg/day in 6-week cycles (4 weeks on, 2 weeks off)	Metastatic breast cancer previously treated with anthracycline and taxane	ORR was 11% with sunitinib and was slightly higher in the TNBC subgroup.
Barrios et al. [65]	III /Metastatic	Sunitinib vs capecitabine	Previously treated Her-2 negative breast cancer (>30% of patients were TNBC)	Median DFS was better for capecitabine compared with sunitinib monotherapy (4.2 vs 2.8 months). OS was not different. In conclusion, sunitinib cannot be recommended as monotherapy for patients with metastatic disease.

Continued on next page

Continued from previous page

	Phase/Setting	*Treatment*	*Study population*	*Results/Comments*
Sorafenib				
Baselga et al. [67]	IIb / Metastatic-first or second lines	Sorafenib 400 mg twice daily + capecitabine vs sorafenib + placebo	Metastatic breast cancer (30% patients were TNBC)	Median PFS was significantly better in sorafenib+capecitabine group.
Gradishar et al. [68]	IIb / Metastatic-first-line	Sorafenib 400 mg twice daily + paclitaxel (90 mg/m^2, weekly, 3 weeks on, 1 week off) or placebo + paclitaxel	Locally recurrent or metastatic breast cancer (40% of patients had TNBC)	Significant improvement was obtained in TTP and ORR with sorafenib+paclitaxel combination.
Erlotinib				
Dickler et al. [58]	II / Locally advanced/ Metastatic	Erlotinib 150 mg orally daily	Locally advanced or metastatic breast cancer	One patient in each cohort had a PR. Erlotinib had minimal activity in unselected previously treated women with advanced breast cancer.
PARP inhibitors				
Iniparib				
O'Shaughnessy et al. [76]	II / Metastatic	Gemcitabine + Carboplatin ± iniparib	Metastatic TNBC	CBR was 56% in iniparib arm vs 34% in chemotherapy arm. PFS and OS rates were significantly better for iniparib arm than chemotherapy (5.9 vs 3.6 months and 12.3 vs 7.7 months, respectively).
O'Shaughnessy et al. [77]	III / Metastatic	Gemcitabine + Carboplatin ± Iniparib	Metastatic TNBC	In the preliminary analysis, the addition of iniparib to chemotherapy did not meet the pre-specified criteria for significance for co-primary endpoints of OS and PFS.
Olaparib				
Tutt et al. [78]	II / Metastatic	Cohort A: Olaparib 400 mg twice daily; Cohort B: Olaparib 100 mg twice daily	Stage IIIB/IIIC or IV breast cancer patients who carried a BRCA mutation; ≥1 prior chemotherapy	ORR was 41% in cohort A and 22% in cohort B. 54% of TNBC patients in cohort A achieved ORR vs 25% in cohort B. PFS was 5.7 vs 3.8 months in cohorts, respectively.
Dent et al. [80]	I / Metastatic	Olaparib 200 mg twice daily + paclitaxel weekly for 3 of 4	Metastatic TNBC	There was a high incidence of neutropenia, leading to reduced paclitaxel dose intensity despite prophylaxis with growth factor support.
Veliparib				
Isakoff et al. [81]	II / Metastatic	Veliparib 40 mg twice daily, days 1-7, temozolomide 150 mg/m^2, days 1-5, cycle 28 days	Metastatic breast cancer	This combination was limited to patients with a BRCA1 mutation (one partial response) and a BRCA2 mutation (one complete and one partial response). Stable disease was seen in 4 patients (>4 months), 2 of whom were BRCA2 deficient.
mTOR inhibitors				
Everolimus				
Ellard et al. [84]	II / Metastatic	Everolimus 10 mg daily vs 70 mg weekly	Metastatic breast cancer	ORR was better in daily group than weekly group (12 vs 0%). Everolimus had activity in metastatic breast cancer dependent on schedule.
Gonzalez-Angulo et al. [85]	II / Neoadjuvant	FEC→paclitaxel vs FEC→paclitaxel + RAD001 (everolimus)	TNBC	The addition of RAD001 to paclitaxel plus FEC in the neoadjuvant setting was associated with a higher 12-week response rate than combination chemotherapy (47.8 vs. 29.6%).

C: cyclophosphamide, CBR: clinical benefit rate, DFS: disease-free survival, E: epirubicin, F: 5-fluorouracil, ORR: overall response rate, OS: overall survival, PR: partial response, pCR: pathological complete response, PFS: progression-free survival, TTP: time to progression, TNBC: triple-negative breast cancer

rapidly with a median PFS of 2 months [54]. The combination regimen was well tolerated, with grade 3 toxicity consisting of rash, fatigue, nausea, and vomiting, which occurred in 6% of the patients. Another phase II study randomly evaluated the effect of carboplatin and weekly irinotecan with or without cetuximab in 165 metastatic breast cancer patients. In the TNBC subgroup (n=72), a higher response rate was achieved with the addition of cetuximab to the treatment regimen (49 vs. 30%) [55]. In these studies, the expression of EGFR was not evaluated. Very recently, another phase II randomized BALI-1 trial was presented at the 2011 San Antonio Breast Cancer Symposium [56]. This study showed that adding cetuximab to cisplatin increased the ORR compared to cisplatin alone (20 vs. 10.3%) and improved the PFS in patients with metastatic TNBC (3.7 vs. 1.5 months). Several trials are currently evaluating the efficacy of adding cetuximab to the chemotherapy regimen in both neoadjuvant and metastatic settings.

Erlotinib is an orally active EGFR tyrosine kinase inhibitor (TKI). Erlotinib in combination with capecitabine/docetaxel chemotherapy has shown activity in metastatic breast cancer [57]. In a phase II study carried out by Dickler et al., a partial response was achieved in 2 out of 69 patients, one of which had triple-negative histology [58]. However, the efficacy of erlotinib remains unclear in the TNBC subgroup.

Another EGFR TKI, gefitinib, showed greater activity in combination with carboplatin and docetaxel than the chemotherapy agents alone [59], but further trials are needed to determine the benefit of gefitinib in the subgroup of TNBC patients. Panitumumab is an antibody that targets EGFR and is being evaluated in patients with TNBC. The results of the preliminary analysis of a neoadjuvant pilot phase II trial were presented at the 2011 ASCO annual meeting. Fifty-eight patients with operable TNBC were prospectively included and this study showed that panitumumab in combination with 5-fluorouracil, epidoxorubicin, and cyclophosphamide (FEC 100) followed by docetaxel resulted in pCR in 17 patients with an overall clinical RR of 80% (47% CR). Conservative surgery was performed in 87% of the patients. This systemic treatment was well tolerated and the main side-effect was skin toxicity [60].

Tyrosine kinase inhibitors

Sunitinib is an oral TKI that inhibits VEGF 1, 2 and 3, platelet-derived growth factor receptor (PDGFR) alpha and beta, c-KIT and colony-stimulating factor 1 [61-63]. In several preclinical studies with breast cancer models, sunitinib showed antitumor activity. In a recent, multicenter, phase II study, Burstein et al. analyzed 64 metastatic breast cancer patients (20 with triple negative tumors) who were previously treated with anthracyclines and taxanes. The authors reported an ORR of 11% with sunitinib. The response rate was slightly higher in the TNBC subgroup (15%; 3 out of 20 patients) [64]. The most common grade 3 non-hematological adverse events included fatigue and hand-foot syndrome. A phase III randomized study (SUN 1107) that evaluated sunitinib vs. capecitabine in patients with previously treated HER-2-negative advanced breast cancer was presented at the 2009 San Antonio Breast Cancer Symposium [65]. More than 30% of the patients had TNBC and less than 2 prior regimens for metastatic disease. The primary end point (DFS) was not reached; indeed, the median DFS was better with capecitabine therapy (4.2 vs. 2.8 months). No significant difference was detected with respect to OS. After these results, the Independent Data Monitoring Committee recommended that trial enrollment be stopped due to futility and concluded that sunitinib cannot be recommended as monotherapy at this dosing schedule for the treatment of advanced metastatic breast cancer. Sunitinib is being evaluated in a phase II study on previously treated patients with metastatic TNBC (NCT00246571). This trial recently completed accrual and the results are eagerly awaited [66]. Another neoadjuvant trial of sunitinib in combination with paclitaxel and carboplatin in locally advanced TNBC is ongoing (NCT00887575).

Sorafenib is another multitargeted TKI with antiangiogenic and antiproliferative activity. It is currently being used for the treatment of patients with advanced renal cell carcinoma and unresectable hepatocellular carcinoma. Sorafenib has shown modest activity as a single agent in advanced breast cancer patients and is currently being evaluated in 2 phase IIB trials. The SOLTI-0701 study evaluated the combination of sorafenib (400 mg twice daily) with capecitabine or a placebo in patients with metastatic breast cancer as first- or second-line treatment. Thirty percent of the patients had TNBC. The median PFS was significantly better for patients treated with the combination of sorafenib-capecitabine than for the patients who received sorafenib-placebo (hazard ratio [HR], 0.57, p=0.0006). Grade 3 hand-foot syndrome was high in the sorafenib-capecitabine arm (45 vs. 13%) [67]. A second study, a phase IIb study, was also presented at the 2009 San Antonio Breast Cancer Symposium. This trial investigated the efficacy of sorafenib in combination with paclitaxel or a placebo as first-line therapy in patients with locally recurrent or metastatic breast cancer. Forty of the patients had TNBC. The results of the primary endpoint, PFS, demonstrated a trend favoring sorafenib over paclitaxel-placebo (HR 0.788, p=0.0857). Moreover, significant improvements in time

to progression and the ORR were observed. The rate of grade 3/4 hand-foot reactions was 30 vs. 3% in the placebo group. From these data, the authors concluded that sorafenib provides added benefits when combined with paclitaxel compared to single-agent paclitaxel in the first-line treatment of advanced breast cancer [68].

The src tyrosine kinase (Rous sarcoma virus) is overexpressed in breast cancer and it may be another potential target for the treatment of TNBC. It was found to be related to metastatic disease progression [69]. Dasatinib is an orally active, small-molecule TKI that acts on src and brc protein kinases, in addition to inhibiting c-kit. Preclinical trials showed that the growth of basal-like breast cancer cell lines was highly sensitive to inhibition by dasatinib [70,71]. A phase II study (BMS-354825 NCT00371254) with dasatinib in patients with advanced TNBC has been recently completed patient recruitment and the results are awaited [72].

Poly-(ADP-ribose)-polymerase inhibitors

Poly-(ADP-ribose)-polymerase (PARP) is a nuclear protein that is recruited to the site of stranded DNA breaks. PARP, especially PARP 1, plays a greater role, together with other mechanisms that include BRCA1 and BRCA2 proteins, in DNA repair when the preferred homologous recombination mechanism for repairing double-stranded breaks is lost because of a BRCA1 dysfunction. This enzyme rescues tumor cells from DNA damage and it may be a good therapeutic target in tumors with a BRCA mutation. In preclinical studies, cancer cells with mutated BRCA1 were found to be hypersensitive to PARP inhibition [73,74]. Molecular defects that bring about BRCA1-related breast cancers also emerge in TNBC. Therefore, PARP inhibitors, either as monotherapy or in combination with chemotherapeutic agents that result in DNA breaks, such as alkylating agents and topoisomerase I inhibitors, could possibly be used in patients with TNBC [75].

Currently, several PARP inhibitors, including iniparib (previously known as BSI-201), olaparib (previously known as AZD2281), and veliparib (previously known as ABT-888) are being evaluated. Iniparib was evaluated in combination with gemcitabine and carboplatin in patients with TNBC. The final results of this phase II trial revealed that the addition of iniparib to gemcitabine and carboplatin significantly improved the clinical benefit rate (56 vs. 34%, p=0.01), PFS (5.9 vs. 3.6 months, p=0.01) and the median OS (12.3 vs. 7.7 months, p=0.01) compared to chemotherapy alone. Iniparib was well tolerated and did not significantly increase toxicity [76]. A confirmatory phase III study using the same regimen was recently presented at the 2011 ASCO annual meeting. In the preliminary analysis, although this study showed a compatible safety profile with that of the phase II trial, the addition of iniparib to gemcitabine and carboplatin did not meet the pre-specified criteria for significance for co-primary endpoints of OS and PFS in patients with metastatic TNBC [77]. Analyses for further clarification of these results are ongoing. Iniparib is also being investigated in 2 neoadjuvant trials (NCT00813956 and the SOLTI NEOPARP study, and NCT01204125, respectively).

Another oral PARP inhibitor is olaparib, which was evaluated in a phase I study. Fong et al. recently analyzed 60 patients with breast cancer, of whom 22 were BRCA1 or BRCA2 mutation carriers, and 1 patient had a strong family history of BRCA-associated cancer. Of the 9 breast cancer patients, 2 BRCA2 mutation carriers achieved a clinical response (one with CR and the other stable disease for 7 months). Patients without the mutation did not show response [73]. Following this, olaparib was evaluated in a phase II trial involving 54 patients with chemotherapy-refractory advanced breast cancer who carried a BRCA1 or BRCA2 mutation. The first cohort (n=27) (13 out of 27 had TNBC) was given continuous oral olaparib at the maximum tolerated dose (400 mg twice daily), and the second (n=27) (16 out of 27 had TNBC) was given a lower dose (100 mg twice daily). An ORR was obtained in 11 (41%) out of 27 patients in the cohort receiving 400 mg twice daily, while 6 (22%) out of 27 patients had an objective response in the 100 mg twice daily cohort. Overall response of the patients with TNBC was 7 (54%) out of 13 in the 400 mg cohort and 4 (25%) out of 16 in the 100 mg cohort. None of the patients who had TNBC achieved pCR. The median PFS was 5.7 months for patients treated with 400 mg and 3.8 months for patients in the 100 mg cohort. This agent was fairly well tolerated, with fatigue and nausea being the most common adverse events in both treatment cohorts [78]. In another smaller phase II study, 400 mg olaparib monotherapy showed an ORR of 0% in 15 patients with TNBC [79]. Recently, Dent et al. presented their results of a phase I study of olaparib in combination with weekly paclitaxel at the 2010 ASCO annual meeting [80]. Although responses were observed with this combination, there was a high incidence of neutropenia, leading to reduced paclitaxel dose intensity despite prophylaxis with G-CSF support. Several clinical trials using olaparib alone or in combination with various chemotherapeutic agents are also under way to evaluate them in TNBC with BRCA-deficient cancers.

Veliparib is another oral PARP1 inhibitor that is being investigated in combination with temozolomide. The results of a phase II trial were recently reported and the activity of this combination was limited to pa-

tients who had BRCA1 mutation (one partial response) and a BRCA2-mutation (one complete and one partial response). Stable disease was seen in 4 patients (>4 months), 2 of whom were BRCA2-deficient. The median PFS was 1.9 months in all patients and 5.5 months in those with BRCA mutations [81].

The novel intravenous PARP inhibitor, PF-01367338, in combination with cisplatin for TNBC patients with residual invasive disease after standard preoperative anthracycline and/or taxane-containing chemotherapy is being evaluated in a phase II study (NCT01074970).

Mammalian target of rapamycin inhibitors

The mammalian target of rapamycin (mTOR) is a cell-cycle regulator protein that is downstream of the PI3K/AKT pathway and, when activated, promotes protein synthesis, cell-cycle progression, proliferation, and angiogenesis [82]. Activation of the PI3K pathway frequently occurs in TNBC and confers susceptibility to mTOR inhibitors. Moreover, loss of the PTEN tumor suppressor gene is common in TNBC, which gives rise to increased mTOR activation [83]. Everolimus, an oral mTOR inhibitor was evaluated in phase II trials as a first- or second-line treatment in 59 metastatic breast cancer patients, of whom 20 were HER-2 negative. A 10 mg daily regimen of single-agent everolimus showed a 12% ORR in heavily pretreated patients with metastatic breast cancer, but the ORR was 0% for the weekly regimen [84]. Very recently, a phase II randomized clinical trial of standard neoadjuvant chemotherapy with paclitaxel followed by FEC vs. the combination of paclitaxel and RAD001 followed by FEC in patients with TNBC was presented at the 2011 ASCO annual meeting [85]. The clinical endpoints were the 12-week response rate, the pCR, and toxicity. Results of the preliminary analysis showed that the addition of RAD001 to paclitaxel plus FEC in the neoadjuvant setting was well tolerated and associated with a higher 12-week response rate than the combination chemotherapy alone (47.8 vs. 29.6%). However, this difference was not statistically significant due to the small sample size. Biomarker analysis is ongoing in order to further optimize patient selection.

Therapy based on androgen receptor inhibition

Preclinical *in vitro* studies indicated that androgens can induce proliferative changes in breast cancer cell lines and promote tumorigenesis in animal models by androgen receptor (AR) stimulation [86]. Doane et al. conducted a genome-wide gene expression profiling study of 99 patients with breast cancer, 41 of whom had TNBC. They found that the androgen-enhanced growth of this breast cancer cell line was ER independent and AR dependent [87]. These authors subsequently identified MDA-MB-453 as a cell line that had a molecular phenotype similar to a previously defined subtype of TNBC. Previous studies reported that 10-35% of TNBC express AR [88,89]. Thus, patients with TNBC may benefit from inhibition of the AR. Bicalutamide, a non-steroidal competitive androgen inhibitor, is used in the treatment of advanced prostate cancer, but it is currently being tested in the treatment of AR-positive TNBC by a multicenter phase II trial (NCT00468715, TBCRC 011study).

Heat shock protein 90 inhibitors

Heat shock proteins (Hsp) are highly conserved proteins and their expression is dependent on the level of various cellular stresses. The Hsp 90 proteins play a role as molecular chaperones for several cellular proteins in transducing proliferative signals. Their function is essential for normal cell viability and growth [90]. Several initial clinical studies showed promising anticancer activity of Hsp 90 inhibitors, mainly in breast cancer and various hematological malignancies. When the function of Hsp 90 is blocked, its client protein is degraded by proteasomes. Geldanamycin and tanespimycin have shown antitumor activity in HER-2-positive metastatic breast cancer patients [91,92]. Clinical trials are evaluating the efficacy of Hsp 90 inhibitors AUY922 and IPI-504, but only in ER and HER-2-positive disease (NCT0181613 and NCT01081600). Whether or not the agents of this class will prove effective *in vivo* and specifically in TNBC remains to be seen.

Histone deacetylase inhibitors

The interaction between histone acetyl transferase and histone deacetylase (HDAC) enzymes modulates the structure of chromatin and transcription factor accessibility, resulting in changes in gene expression. Inhibitors of HDAC have pleiotropic effects on cell cycle arrest, apoptosis, and differentiation, and the inhibition of growth and angiogenesis have emerged as promising new therapeutic agents in multiple cancers, including those resistant to standard chemotherapy. HDAC inhibitor activity has been found in breast cancer cell lines in *in vitro* studies [93,94]. Various HDAC inhibitors, such as trichostatin A and SK-7041, have been used in preclinical trials, and vorinostat has been used in phase I and II studies [95,96]. Recently, a new HDAC inhibitor, entinostat (MS-275), in combination with all trans retinoic acid (ATRA) and low dose chemotherapy, resulted in the regression of established xenografts of TNBC [97].

Tumor necrosis factor-related apoptosis-inducing ligand inhibitors

The tumor necrosis factor (TNF)-alpha-related apoptosis-inducing ligand (TRAIL) recently emerged as a promising targeted therapeutic strategy in various types of cancers due to its proapoptotic characteristics. As a member of the TNF superfamily, TRAIL specifically activates extrinsic apoptotic pathways in cancer cells by binding to death receptors (DR4 and DR5). Importantly, TRAIL selectively promotes the apoptosis of tumor cells but it has no effect on normal human reproductive tract cells, including those in the endometrium, ovary, cervix, and fallopian tubes [98,99]. Agonistic monoclonal antibodies (mAbs) that functionally engage human and murine TRAIL receptors have shown their capability to engage human DR4 or DR5 in exerting antitumor activity in *in vivo* xenograft studies [100]. Phase II trials are currently investigating the therapeutic efficacy of TRAIL agonists alone or in combination with various chemotherapeutics. Tigatuzumab is a humanized, anti-DR5, agonistic, monoclonal antibody that has strong preclinical data showing its *in vitro* and *in vivo* activity against basal-like breast cancer cells. A phase II trial using nanoparticle-albumin-bound paclitaxel with or without tigatuzumab in patients with metastatic TNBC is currently ongoing (TBCRC 019 trial, NCT01307891).

Other therapies

Vitamin D levels may be a risk or an important prognostic factor in breast cancer patients. Many epidemiologic, preclinical and clinical trials have indicated that vitamin D deficiency may result in breast cancer initiation and progression. However, the significance of vitamin D deficiency has not been studied comprehensively in different breast cancer phenotypes. Rainville et al. showed that in their case series TNBC phenotype had the lowest average vitamin D level and the highest percentage of patients that were vitamin D deficient [101]. In addition, checkpoint kinase 1(CHK1) and claspin were quickly down-regulated in mouse MC3T3-E1 mammary carcinoma cells by treatment with 1,25 dihydroxyvitamin D3, which is a known inhibitor of cell proliferation [102]. These findings suggest that low vitamin D levels may be characteristic of TNBC and vitamin D intake may be effective in the treatment of TNBC [103]. On the other hand, the recommendation of routine vitamin D supplementation should be confirmed in large prospective studies for patients with breast cancer including TNBC phenotype.

Conclusion

TNBCs comprise a heterogeneous breast cancer subgroup that substantially overlaps basal-like tumors in terms of their characteristics. However, the biological aspects of TNBC, basal-like breast cancer and BRCA-deficient tumors are specific and different. TNBC is characterized by an aggressive clinical course and poor survival, but only few developments have been made for patients with TNBC. While conventional chemotherapeutic regimens can be successful in treating women with TNBC and basal-like disease, emerging therapies aimed at damaging DNA, angiogenic players, tubulin structures, mTOR, TRAIL, AR, and Hsp 90 have shown promise in early studies but their clinical performance has yet to be definitively proven. Novel therapeutic options will be provided for best approaching the treatment of TNBCs. Future studies are needed for the detection of novel molecular predictive factors and therapeutic targets in order to identify better and optimal treatment modalities for TNBC.

References

1. Brenton JD, Carey LA, Ahmed AA, Caldas C. Molecular classification and molecular forecasting of breast cancer: ready for clinical application? J Clin Oncol 2005; 23: 7350-7360.
2. Perou CM, Sørlie T, Eisen MB et al. Molecular portraits of human breast tumours. Nature 2000; 406: 747-752.
3. Rakha EA, Reis-Filho JS, Ellis IO. Basal-like breast cancer: a critical review. J Clin Oncol 2008; 26: 2568-2581.
4. Nielsen TO, Hsu FD, Jensen K et al. Immunohistochemical and clinical characterization of the basal-like subtype of invasive breast carcinoma. Clin Cancer Res 2004; 10: 5367-5374.
5. Dent R, Trudeau M, Pritchard KI et al. Triple-negative breast cancer: clinical features and patterns of recurrence. Clin Cancer Res 2007; 13: 4429-4434.
6. Kaplan HG, Malmgren JA, Atwood M. T1N0 Triple Negative Breast Cancer: Risk of Recurrence and Adjuvant Chemotherapy. Breast J 2009; 15: 454-460.
7. Bauer KR, Brown M, Cress RD, Parise CA, Caggiano V. Descriptive analysis of estrogen receptor (ER)-negative, progesterone receptor (PR)-negative, and HER2-negative invasive breast cancer, the so-called triple-negative phenotype: a population-based study from the California Cancer Registry. Cancer 2007; 109: 1721-1728.
8. Carey LA, Perou CM, Livasy CA et al. Race, breast cancer subtypes, and survival in the Carolina Breast Cancer Study. JAMA 2006; 295: 2492-2502.
9. Aksoy S, Dizdar O, Harputluoglu H, Altundag K. Demographic, clinical, and pathological characteristics of Turkish triple-negative breast cancer patients: single center experience. Ann Oncol 2007; 18: 1904-1906.
10. Van Calster B, Vanden Bempt I, Drijkoningen M et al. Axillary lymph node status of operable breast cancers by combined steroid receptor and HER-2 status: triple positive tumours are more likely lymph node positive. Breast Cancer Res Treat

2009; 113: 181-187.
11. Narod SA, Foulkes WD. BRCA1 and BRCA2: 1994 and beyond. Nat Rev Cancer 2004; 4: 665-676.
12. Couch FJ. Genetic epidemiology of BRCA1. Cancer Biol Ther 2004; 3: 509-514.
13. Young SR, Pilarski RT, Donenberg T et al. The prevalence of BRCA1 mutations among young women with triple-negative breast cancer. BMC Cancer 2009; 9: 86.
14. Pal SK, Mortimer J. Triple-negative breast cancer: novel therapies and new directions. Maturitas 2009; 63: 269-274.
15. Atchley DP, Albarracin CT, Lopez A et al. Clinical and pathologic characteristics of patients with BRCA-positive and BRCA-negative breast cancer. J Clin Oncol 2008; 26: 4282-4288.
16. Miyoshi Y, Murase K, Oh K. Basal-like subtype and BRCA1 dysfunction in breast cancers. Int J Clin Oncol 2008; 13: 395-400.
17. Turner NC, Reis-Filho JS. Basal-like breast cancer and the BRCA1 phenotype. Oncogene 2006; 25: 5846-5853.
18. Haffty BG, Yang Q, Reiss M et al. Locoregional relapse and distant metastasis in conservatively managed triple negative early-stage breast cancer. J Clin Oncol 2006; 24: 5652-5657.
19. Morris GJ, Naidu S, Topham AK et al. Differences in breast carcinoma characteristics in newly diagnosed African-American and Caucasian patients: a single-institution compilation compared with the National Cancer Institute's Surveillance, Epidemiology, and End Results database. Cancer 2007; 110: 876-884.
20. 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: 1275-1281.
21. Carey LA, Dees EC, Sawyer L et al. The triple negative paradox: primary tumor chemosensitivity of breast cancer subtypes. Clin Cancer Res 2007; 13: 2329-2334.
22. Hayes DF, Thor AD, Dressler LG et al. HER2 and response to paclitaxel in node-positive breast cancer. N Engl J Med 2007; 357: 1496-1506.
23. Ellis P, Barrett-Lee P, Johnson L et al. Sequential docetaxel as adjuvant chemotherapy for early breast cancer (TACT): an open-label, phase III, randomised controlled trial. Lancet 2009; 373: 1681-1692.
24. Rouzier R, Perou CM, Symmans WF et al. Breast cancer molecular subtypes respond differently to preoperative chemotherapy. Clin Cancer Res 2005; 11: 5678-5685.
25. Martin LP, Hamilton TC, Schilder RJ. Platinum resistance: the role of DNA repair pathways. Clin Cancer Res 2008; 14: 1291-1295.
26. Kelland L. The resurgence of platinum-based cancer chemotherapy. Nat Rev Cancer 2007; 7: 573-584.
27. Byrski T, Gronwald J, Huzarski T, Grzybowska E et al. Pathologic complete response rates in young women with BRCA1-positive breast cancers after neoadjuvant chemotherapy. J Clin Oncol 2010; 28: 375-379.
28. Silver DP, Richardson AL, Eklund AC et al. Efficacy of neoadjuvant cisplatin in triple-negative breast cancer. J Clin Oncol 2010; 28: 1145-1153.
29. 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: 667-672.
30. Frasci G, Comella P, Rinaldo M et al. Preoperative weekly cisplatin-epirubicin-paclitaxel with G-CSF support in triple-negative large operable breast cancer. Ann Oncol 2009; 20: 1185-1192.
31. Yi S, Uhm J, Cho E et al. Clinical outcomes of metastatic breast cancer patients with triple-negative phenotype who received platinum-containing chemotherapy. Proc Am Soc Clin Oncol 2008; 26: 15S (abstr 1008).
32. Sirohi B, Arnedos M, Popat S et al. Platinum-based chemotherapy in triple-negative breast cancer. Ann Oncol 2008; 19: 1847-1852.
33. Uhm JE, Park YH, Yi SY et al. Treatment outcomes and clinicopathologic characteristics of triple-negative breast cancer patients who received platinum-containing chemotherapy. Int J Cancer 2009; 124: 1457-1462.
34. Isakoff SJ, Goss PE, Mayer EL et al. TBCRC009: A multicenter phase II study of cisplatin or carboplatin for metastatic triple-negative breast cancer and evaluation of p63/p73 as a biomarker of response. Proc Am Soc Clin Oncol 2011; 29: 15S (abstr 1025).
35. Di Leo A, Isola J, Piette F et al. A meta-analysis of phase III trials evaluating the predictive value of Her-2 and topoisomerase II alpha in early breast cancer patients treated with CMF or anthracycline-based adjuvant therapy. Breast Cancer Res Treat 2008; 107: 24.
36. Hugh J, Hanson J, Cheang MC et al. Breast cancer subtypes and response to docetaxel in node-positive breast cancer: use of an immunohistochemical definition in the BCIRG 001 trial. J Clin Oncol 2009; 27: 1168-1176.
37. Bear HD, Anderson S, Brown A et al. The effect on tumor response of adding sequential preoperative docetaxel to preoperative doxorubicin and cyclophosphamide: preliminary results from National Surgical Adjuvant Breast and Bowel Project Protocol B-27. J Clin Oncol 2003; 21: 4165-4174.
38. Smith IC, Heys SD, Hutcheon AW et al. Neoadjuvant chemotherapy in breast cancer: significantly enhanced response with docetaxel. J Clin Oncol 2002; 20: 1456-1466.
39. Sanchez-Munoz A, Garcia-Tapiador AM, Martinez-Ortega E et al. Tumour molecular subtyping according to hormone receptors and HER2 status defines different pathological complete response to neoadjuvant chemotherapy in patients with locally advanced breast cancer. Clin Transl Oncol 2008; 10: 646-653.
40. Knoop AS, Knudsen H, Balslev E et al. Retrospective analysis of topoisomerase IIa amplifications and deletions as predictive markers in primary breast cancer patients randomly assigned to cyclophosphamide, methotrexate, and fluorouracil or cyclophosphamide, epirubicin, and fluorouracil: Danish Breast Cancer Cooperative Group. J Clin Oncol 2005; 23: 7483-7490.
41. Tanner M, Isola J, Wiklund T et al. Topoisomerase IIalpha gene amplification predicts favorable treatment response to tailored and dose-escalated anthracycline-based adjuvant chemotherapy in HER-2/neu-amplified breast cancer: Scandinavian Breast Group Trial 9401. J Clin Oncol 2006; 24: 2428-2436.
42. Bond GL, Hu W, Bond EE et al. A single nucleotide polymorphism in the MDM2 promoter attenuates the p53 tumor suppressor pathway and accelerates tumor formation in humans. Cell 2004; 119: 591-602.
43. Bidard FC, Matthieu MC, Chollet P et al. p53 status and efficacy of primary anthracyclines/alkylating agent-based regimen according to breast cancer molecular classes. Ann Oncol 2008; 19: 1261-1265.
44. Kavallaris M. Microtubules and resistance to tubulin-binding agents. Nat Rev Cancer 2010; 10: 194-204.
45. Pivot XB, Li RK, Thomas ES et al. Activity of ixabepilone

in oestrogen receptor-negative and oestrogen receptor-progesterone receptor-human epidermal growth factor receptor 2-negative metastatic breast cancer. Eur J Cance. 2009; 45: 2940-2946.
46. Baselga J, Zambetti M, Llombart-Cussac A et al. Phase II genomics study of ixabepilone as neoadjuvant treatment for breast cancer. J Clin Oncol 2009; 27: 526-534.
47. Thomas ES, Gomez HL, Li RK et al. Ixabepilone plus capecitabine for metastatic breast cancer progressing after anthracyclines and taxane treatment. J Clin Oncol 2007; 25: 5210-5217.
48. Roche H, Li RK, Ro J. Ixabepilone plus capecitabine improves progression free survival in patients with metastatic breast cancer resistant to taxanes: a pooled analysis from two phase III trials. Cancer Res 2009; 69: S2 (abstr 2015).
49. Miller K, Wang M, Gralow J et al. Paclitaxel plus bevacizumab versus paclitaxel alone for metastatic breast cancer. N Engl J Med 2007; 357: 2666-2676.
50. Miles D, Chan A, Romieu G. Randomized, double-blind, placebo-controlled, phase III study of bevacizumab with docetaxel or docetaxel with placebo as first-line therapy for patients with locally recurrent or metastatic breast cancer (mBC): AVADO. Proc Am Soc Clin Oncol 2008; 26: 15S (abstr 1011).
51. Robert N, Dieras V, Glaspy J. Ribbon-1: randomized, double-blind, placebo-controlled, phase III trial of chemotherapy with or without bevacizumab (B) for first-line treatment of HER2-negative locally recurrent or metastatic breast cancer (MBC). Proc Am Soc Clin Oncol 2009; 27: 15S (abstr 1005).
52. O'Shaughnessy J, Dieras V, Glaspy J et al. Comparison of Subgroup Analyses of PFS from Three Phase III Studies of Bevacizumab in Combination with Chemotherapy in Patients with HER2-Negative Metastatic Breast Cancer (MBC). Cancer Res 2009; 69: S1 (abstr 207)
53. Balduzzi A, Montagna E, Bagnardi V et al. Infusional fluorouracil, epirubicin, and cisplatin followed by weekly paclitaxel plus bevacizumab in locally advanced breast cancer with unfavorable prognostic features. Anticancer Drugs 2009; 20: 197-203.
54. Carey LA, Rugo HS, Marcom PK et al. TBCRC 001: EGFR inhibition with cetuximab added to carboplatin in metastatic triple-negative (basal-like) breast cancer. Proc Am Soc Clin Oncol 2008; 26: 15S (abstr 1009).
55. O'Shaughnessy J, Weckstein DJ, Vukelja SJ et al. Preliminary results of a randomized phase II study of weekly irinotecan/carboplatin with or without cetuximab in patients with metastatic breast cancer. Breast Cancer Res Treat 2007; 106: S32 (abstr 308).
56. Baselga J, Stemmer S, Pego A et al. Cetuximab + Cisplatin in Estrogen Receptor-Negative, Progesterone Receptor-Negative, HER2-Negative (Triple-Negative) Metastatic Breast Cancer: Results of the Randomized Phase II BALI-1 Trial. Cancer Res 2010; 70: 24S2 (abstr PD01-01).
57. Twelves C, Trigo JM, Jones R et al. Erlotinib in combination with capecitabine and docetaxel in patients with metastatic breast cancer: a dose-escalation study. Eur J Cancer 2008; 44: 419-426.
58. Dickler MN, Cobleigh MA, Miller KD et al. Efficacy and safety of erlotinib in patients with locally advanced or metastatic breast cancer. Breast Cancer Res Treat 2009; 115: 115-121.
59. Corkery B, Crown J, Clynes M, O'Donovan N. Epidermal growth factor receptor as a potential therapeutic target in triple-negative breast cancer. Ann Oncol 2009; 20: 862-867.
60. Nabholtz J, Weber B, Mouret-Reynier M et al. Panitumumab in combination with FEC 100 (5-fluorouracil, epidoxorubicin, cyclophosphamide) followed by docetaxel (T) in patients with operable, triple-negative breast cancer (TNBC): Preliminary results of a multicenter neoadjuvant pilot phase II study. Proc Am Soc Clin Oncol 2011; 29: 15S (abstr e11574).
61. Mendel DB, Laird AD, Xin X et al. In vivo antitumor activity of SU11248, a novel tyrosine kinase inhibitor targeting vascular endothelial growth factor and platelet derived growth factor receptors: determination of a pharmacokinetic/ pharmacodynamic relationship. Clin Cancer Res 2003; 9: 327-337.
62. Abrams TJ, Lee LB, Murray LJ et al. SU 11248 inhibits KIT and platelet-derived growth factor receptor beta in preclinical models of human small cell lung cancer. Mol Cancer Ther 2003; 2: 757-766.
63. Deeks ED, Keating GM. Sunitinib. Drugs 2006; 66: 2255-2266; discussion 67-68.
64. Burstein HJ, Elias AD, Rugo HS et al. Phase II study of sunitinib malate, an oral multitargeted tyrosine kinase inhibitor, in patients with metastatic breast cancer previously treated with an anthracycline and a taxane. J Clin Oncol 2008; 26: 1810-1816.
65. Barrios C, Liu M, Lee S et al. Phase III randomized trial of sunitinib versus capecitabine in patients with previously treated HER 2 negative advanced breast disease (SUN 1107) Cancer Res 2009; 69: 24S1(abstr 46).
66. Study of SU011248 versus chemotherapy for patients with previously treated triple receptor negative breast cancer. Available from: www.clinicaltrials.gov [Last accessed 26 August 2011].
67. Baselga J, Roche H, Costa F et al. SOLTI 0701: a multinational double-blind, randomized phase 2b study evaluating the efficacy and safety of sorafenib compared to placebo when administered in combination with capecitabine in patients with locally advanced or metastatic breast cancer Cancer Res 2009; 69: 24S1 (abstr 45).
68. Gradishar W, Kaklamani V, Prasad Sahoo T et al. A double-blind, randomized, placebo-controlled, phase 2b study evaluating the efficacy and safety of sorafenib in combination with paclitaxel as a first line therapy in patients with locally recurrent or metastatic breast cancer. Cancer Res 2009; 69: 24S1 (abstr 44).
69. Conlin AK, Seidman AD. Beyond cytotoxic chemotherapy for the first line treatment of HER 2-negative, hormone insensitive metastatic breast cancer: current status and future opportunities. Clin Breast Cancer 2008; 8: 215-223.
70. Finn RS, Dering J, Ginther C et al. Dasatinib, an orally active small molecule inhibitor of both the src and abl kinases, selectively inhibits growth of basal-type/"triple-negative" breast cancer cell lines growing in vitro. Breast Cancer Res Treat 2007; 105: 319-326.
71. Verbeek B, Vroom T, Adriansen-Slot S et al. c-Src protein expression is increased in human breast cancer. An immunohistochemical and biochemical analysis. J Pathol 1996; 180: 383-388.
72. Phase II Study of Dasatinib (BMS-354825) for Advanced "Triple-Negative" Breast Cancer. Available from: www.clinicaltrials.gov [Last accessed 15 May 2009].
73. Fong PC, Boss DS, Yap TA et al. Inhibition of poly(ADP-ribose) polymerase in tumors from BRCA mutation carriers. N Engl J Med 2009; 361: 123-134.
74. McCabe N, Turner NC, Lord CJ et al. Deficiency in the repair of DNA damage by homologous recombination and sensitivity to poly(ADP-ribose) polymerase inhibition. Cancer Res 2006; 66: 8109-8115.

75. Sahin I, Ararat E, Altundag K. BRCA-deficient and triple negative breast cancers: is olaparib effective in both subtypes? J BUON 2011; 16: 184-185.
76. O'Shaughnessy J, Osborne C, Pippen JE et al. Iniparib plus chemotherapy in metastatic triple-negative breast cancer. N Engl J Med 2011; 364: 205-214.
77. O'Shaughnessy J, Schwartzberg LS, Danso MA et al. A randomized phase III study of iniparib (BSI-201) in combination with gemcitabine/carboplatin (G/C) in metastatic triple-negative breast cancer (TNBC). Proc Am Soc Clin Oncol 2011; 29: 15S (abstr 1007).
78. Tutt A, Robson M, Garber JE et al. Oral poly(ADP-ribose) polymerase inhibitor olaparib in patients with BRCA1 or BRCA2 mutations and advanced breast cancer: a proof-of-concept trial. Lancet 2010; 376: 235-244.
79. Gelmon KA, Hirte HW, Robidoux A et al. Can we define tumors that will respond to PARP inhibitors? A phase II correlative study of olaparib in advanced serous ovarian cancer and triple-negative breast cancer. Proc Am Soc Clin Oncol 2010; 28: 15S (abstr 3002).
80. Dent R, Lindeman G, Clemons M et al. Safety and efficacy of the oral PARP inhibitor olaparib (AZD2281) in combination with paclitaxel for the first or second-line treatment of patients with metastatic triple-negative breast cancer: Results from the safety cohort of a phase I/II multicenter trial. Proc Am Soc Clin Oncol 2010; 28: 15S (abstr 1018).
81. Isakoff SJ, Overmoyer B, Tung NM et al. A phase II trial of the PARP inhibitor veliparib (ABT888) and temozolomide for metastatic breast cancer. Proc Am Soc Clin Oncol 2010; 28: 15S (abstr 1019).
82. Meric-Bernstam F, Gonzalez-Angulo AM. Targeting the mTOR signaling network for cancer therapy. J Clin Oncol 2009; 27: 2278-2287.
83. Saal LH, Holm K, Maurer M et al. PIK3CA mutations correlate with hormonoreceptors, node metastasis and ERBB2 and are mutually exclusive with PTEN loss in human breast carcinoma. Cancer Res 2005; 65: 2554-2559.
84. Ellard SL, Clemons M, Gelmon KA et al. Randomized phase II study comparing two schedules of everolimus in patients with recurrent/metastatic breast cancer: NCIC clinical trials group IND.163. J Clin Oncol 2009; 27: 4536-4541.
85. Gonzalez-Angulo AM, Green MC, Murray JL et al. Open label, randomized clinical trial of standard neoadjuvant chemotherapy with paclitaxel followed by FEC (T-FEC) versus the combination of paclitaxel and RAD001 followed by FEC (TR-FEC) in women with triple receptor-negative breast cancer (TNBC). Proc Am Soc Clin Oncol 2011; 29: 15S (abstr 1016).
86. Gucalp A, Traina TA. Triple-negative breast cancer: role of the androgen receptor. Cancer J 2010; 16: 62-65.
87. Doane AS, Danso M, Lal P et al. An estrogen receptor-negative breast cancer subset characterized by a hormonally regulated transcriptional program and response to androgen. Oncogene 2006; 25: 3994-4008.
88. Gonzalez-Angulo AM, Stemke-Hale K, Palla S et al. Androgen receptor levels and association with PIK3CA mutations and prognosis in breast cancer. Clin Cancer Res 2009; 15: 2472-2478.
89. Park S, Koo J, Park HS et al. Expression of androgen receptors in primary breast cancer. Ann Oncol 2009; 21: 488-492.
90. Whitesell L, Mimmaugh E, De Costa B et al. Inhibition of HSP90-pp60v-src heteroprotein complex formation by benzoquinone ansamycins: essential role for stress proteins in oncogenic transformation. Proc Natl Acad Sci U S A 1994; 91: 8324-8328.
91. Modi S. Heat shock protein 90 inhibition: a novel strategy for the treatment of HER2-positive breast cancer. Cancer Res 2009; 24S1.
92. Caldas-Lopez E, Cerchietti L, Ahn J et al. Hsp90 inhibitor PU-H71, a multimodal inhibitor of malignancy induces complete responses in triple negative breast cancer models. Proc Natl Acad Sci U S A 2009; 106: 8368-8373.
93. Bolden JE, Peart MJ, Johnstone RW. Anticancer activities of histone deacetylase inhibitors. Nat Rev Drug Discov 2006; 5: 769-784.
94. Marks PA, Jiang X. Histone deacetylase inhibitors in programmed cell death and cancer therapy. Cell Cycle 2005; 4: 549-551.
95. Luu TH, Morgan RJ, Leong L et al. A phase II trial of vorinostat (suberoylanilide hydroxamic acid) in metastatic breast cancer: a California Cancer Consortium study. Clin Cancer Res 2008; 14: 7138-7142.
96. Lee KW, Kim JH, Park JH et al. Antitumor activity of SK-7041, a novel histone deacetylase inhibitor, in human lung and breast cancer cells. Anticancer Res 2006; 26: 3429-3438.
97. Nguyen NK, Korangath P, Sabnis G et al. A Combination of HDAC Inhibitor Entinostat (MS-275), All Trans Retinoic Acid (ATRA) and Chemotherapy Drug(s) Causes Regression of Established Xenografts of Triple Negative Breast Cancer. Cancer Res 2010; 70: 24S2 (abstr PD01-05).
98. Holoch PA, Griffith TS. TNF-related apoptosis-inducing ligand (TRAIL): a new path to anti-cancer therapies. Eur J Pharmacol 2009; 625: 63-72.
99. Sadarangani A, Kato S, Espinoza N et al. TRAIL mediates apoptosis in cancerous but not normal primary cultured cells of the human reproductive tract. Apoptosis 2007; 12: 73-85.
100. Cretney E, Takeda K, Smyth MJ. Cancer: novel therapeutic strategies that exploit the TNF-related apoptosis-inducing ligand (TRAIL)/TRAIL receptor pathway. Int J Biochem Cell Biol 2007; 39: 280-286.
101. Rainville C, Khan Y, Tisman G. Triple negative breast cancer patients presenting with low serum vitamin D levels: a case series. Cases J 2009; 2; 8390.
102. Verlinden L, Eelen G, Van Hellemont R et al. 1a,25-Dihydroxyvitamin D(3)-induced down regulation of the checkpoint proteins, Chk1 and claspin, is mediated by the pocket proteins p107 and p130. J Steroid Biochem Mol Biol 2007; 103: 411-415.
103. 103. Harputluoglu H, Dizdar O, Karaahmet F, Altundag K. Vitamin D intake may be effective in the management of triple-negative breast cancer. J BUON 2011; 16: 569.