COMMENTARY

Triple Negative Breast Cancer: Therapeutic and Prognostic Implications

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Abstract

Triple negative breast cancers (TNBC) lack oestrogen receptor(ER), progesterone receptor (PR), nor over-express human epidermal growth factor receptor 2 (HER2). Epidemiologic studies demonstrate that women diagnosed with TNBC manifest a significantly different set of clinic-pathologic features and risk factors when compared to women with other subtypes of breast cancer. They are associated with poor prognosis, as defined by low five-year survival. To date many studies have examined the utility of traditional chemotherapy for the treatment of patients with TNBC and have confirmed the benefits of these agents in both the adjuvant and neoadjuvant settings. Targeted therapy options involving PARP1 and EGFR inhibition, are currently in different phases of development and will hopefully change the paradigm of how patients with TNBC are treated. The present commentary aims to summarize the latest findings on chemotherapy in the treatment of TNBC in both the neoadjuvant and adjuvant setting and explore the ongoing development of newer targeted agents.

Keywords: Triple-negative breast cancer - chemotherapy - epidemiology - prognosis - targeted agents.

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Introduction

The ducts and lobules of normal breast tissue are lined by two distinct layers of cells; the inner or luminal layer and an outer layer which is closer to the basement membrane. These two populations are immunophenotypically distinct (Böcker et al., 1992). Generally luminal cells express low molecular weight cytokeratins as well as oestrogen and progesterone receptors (CK 7,8,18,19; BCL2; ER/PR increased and CK 5,14,17;HER 2 decreased). The basally located cells are a heterogenous population with features of both myoepithelial and smooth muscle cells (CK 5,14,17 ; EGFR increased and ER/PR, HER2 decreased). The myoepithelial cells express high molecular weight cytokeratins and are negative for hormone receptors.

Molecular profiling by modern genomic techniques has identified different subtypes of breast cancer (Table 1). These subtypes show distinct epidemiologic, anthropometric and reproductive associations that possibly indicate different pathways of development. The luminal subtypes (A and B) express hormone receptor related genes and broadly speaking show better outcomes. The hormone receptor negative subtypes include the "basal-like" variant which is characterized by absence or low expression of oestrogen receptor (ER), lack of over-expression of human epidermal growth factor receptor 2 (HER2) and the expression of genes normally associated with the basal cells of the normal breast (Perou et al., 2000). The luminal A and basal-like subtypes have been most accurately distinguished, and are associated with significant variability in clinical outcomes; patients with the latter experience shorter overall and disease free survival. Differences in pathologic responses to chemotherapy have also been reported.

While not synonymous, there is significant overlap between the entities "basal-like" and "triple negative" breast cancer. The latter phenotype is a heterogeneous group defined clinically by negative staining for ER, PR, and HER2 on immunohistochemistry, accounts for 12 to 17 % of breast cancers, and has demonstrated distinct clinical and pathological features with implications for therapy and prognosis (Carey et al., 2007; Rakha et al., 2007). About 90% of triple negative breast cancers (TNBC) tumors fall within the basal-like subgroup, so called for its gene expression profile that mimics basal epithelial cells in other parts of the body (usually identified by IHC staining for the expression of cytokeratin 5/6, reduced ER/PR, and HER2 expression) and a characteristic morphology that

Table 1. Subtypes of Breast Cancer

Molecular subtype	Equivalent Receptor Status	
	Positive	Negative
Luminal A	ER/PR	HER2
Luminal B	ER/PR ; HER 2	
HER2 positive	HER2	ER/PR
Basal-like*		ER/PR ; HER2

*25% discordance between basal- like molecular profile and triple negative status

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includes high proliferative rate, central necrosis, and a pushing border (Livasy et al., 2006; Kreike et al., 2007) The heterogenous nature of breast cancer was not appreciable with traditional histo-pathological assessment, but has been elucidated by gene-expression analysis. In the same way that the impact of hormonal and later trastuzumab therapy justified the distinction between ER and HER-2 negative and positive cancers respectively, there are likely therapeutic implications for the subtypes identified by DNA micro-array based studies. Molecular markers are likely to play an increasing role as targets for the systemic therapy of breast cancer.

Triple negative breast cancer has attracted significant research interest over the last five years (Foulkes et al., 2010). Several issues regarding the precise definition, epidemiology and response to chemotherapy are still being clarified; however research to date has served to shed some light on their distinct epidemiology, pathology and behavior compared to non-triple negative cancers. Although hormonal therapy and trastuzumab are precluded from the adjuvant or neoadjuvant armamentarium for TNBC, the expression of epidermal growth factor receptor (EGFR) and vascular endothelial growth factor (VEGF) provide interesting possibilities for targeted therapy in these tumours. The present review aims to summarize the latest findings on chemotherapy in the treatment of TNBC in both the neoadjuvant and adjuvant setting and explore the ongoing development of newer targeted agents.

Epidemiology

Several studies have indicated some distinctive epidemiological features of TNBC. Women with these tumours are more likely to be black and pre-menopausal (Bauer et al., 2007; Morris et al., 2007). Additionally, there is significant overlap between BRCA1 associated and TNBC - particularly in younger women (Shaheenah, 2010). Comm,on features include high grade, EGFR over expression, P 53 mutations, C-myc amplififcation and cytogenetic abnormalities. Risk factors associated with TNBC -in contrast to hormone receptor positive tumors include younger age at menarche, higher parity, earlier age at first full term pregnancy, and shorter duration of breast feeding (Lin et al., 2009; Harris et al., 2006). They are also associated with higher body mass index (BMI) and waist to hip ratio (WHR) in pre-menopausal women (Millikan et al., 2008). Discrepant results have been reported for the relationship with BMI and WHR; this may be due to differences in study populations, laboratory protocols for assignation of receptor negative status, or the use of different classifications for subtypes. The risk profile for TNBC suggests that prevention strategies in young black females may include life-style modification to decrease abdominal obesity and prolong breast feeding. Further, Dolle et al. (2009) reported that oral contraceptive use >1 year was associated with a 2.7-fold increased risk for TNBC in women under 45 years of age. The risk for triple-negative breast cancer was further heightened in relation to longer oral contraceptive duration and fewer years since last use (Dolle et al., 2009).

Triple negative breast cancers have been associated with several clinico-pathological features that portend increased aggression (Haffty et al., 2006; Carey et al., 2007). These include onset at a younger age, larger size and higher tumour grade, as well as poor correlation between tumour size and nodal involvement. Some studies suggest that the link between tumour size and survival may be more tenuous than in other subtypes (Dent et al., 2007). Mammographic detection is limited by rapid growth and a younger age at presentation and TNBC has been shown to be over-represented among interval breast cancers; on the other hand, when compared to hormone receptor positive counterparts TNBC may show specific features on magnetic resonance imaging.

Prognostic Implications

Triple negative breast cancer is not homogenous with respect to histopathology. The majority of TNBC are invasive ductal carcinomas, however phenotypes such as metaplastic, atypical medullary and adenoid cystic are also seen. Increased tumour size, tumour grade and frequency of nodal involvement have been reported. The common morphological features of TNBC include cellular pleomorphism, high N:C ratio, high mitotic rate, a pushing border of invasion and central or comedo-type necrosis (Carey et al., 2006).

Patients with TNBC are more likely to experience death and distant recurrence compared to those with other cancers, and the median time to death or distant recurrence is significantly shorter; however distant recurrence is less likely to be preceded by a local recurrence. This pattern of behavior may be suggestive of a different method of tumour spread. The elevated risk of death or distant recurrence is sustained for five years from diagnosis, but decreases subsequently and death from TNBC is very unlikely more than ten years after diagnosis (Dent et al., 2007). The sites of distant recurrence are also different than in patients with non-TN cancer, with higher prevalence of visceral rather than skeletal metastases when compared to their hormone receptor-positive counterparts (Liedtke et al., 2008; Smid et al., 2008) and a penchant for lung and brain metastases. Lin et al. (2009) reported that women with TNBC were more likely to develop lung (Odds Ratio (OR) 2.27, $p = 0.0001$) or brain metastases (OR 5.32, $p < 0.0001$) as their first site of recurrence compared with those demonstrated a much lower risk of bone recurrence (OR 0.23, $p < 0.0001$). Studies such as Heitz et al. (2009) demonstrated that women with TNBC have a higher risk for cerebral metastasis compared with patients bearing the ER+/HER2- phenotype and develop cerebral metastases earlier in the course of the disease. Niwinska et al. (2010) reported median survival from brain metastases in triple-negative women of 3.7 months and the authors concluded that survival from brain metastases depended on performance status and the use of systemic treatment. The characteristic behavioural features of triple negative breast cancer are summarized in Table 2.

Therapeutic Implications - Chemotherapy

Triple negative breast cancer is generally associated

Table 2. Prognostic Features of Triple Negative Breast Cancer

Node Positivity with small tumours
High tumour grade
High recurrence rate
"De-Novo" distal recurrence
Short interval from distal recurrence to death
Increased mortality rate up to 5 years
Mortality low after 8 years

with a worse prognosis; nevertheless current data do not imply chemo-resistance of these tumours. Studies done with Anthracycline /Taxane therapy in the neo-adjuvant setting have demonstrated higher pathologic complete response rates (pCR) compared to hormone positive counterparts (Carey et al., 2007). In a prospective study of 1,118 patients treated with neoadjuvant chemotherapy (> 80% treated with anthracycline-based regimen; 53% treated with an additional taxane), patients with TNBC compared with non-TNBC had significantly higher pCR rates (22% v 11%; $p = 0.034$), but decreased 3-year progression-free survival (PFS) rates ($p < 0.0001$) and 3-year overall survival (OS) rates ($p < 0.0001$; Liedtke et al., 2008). A retrospective analysis of 1,731 patients with stage I to III non-inflammatory breast cancer treated predominantly with anthracycline and anthracycline/ taxane containing pre-operative regimens (91% and 58%, respectively), which included 317 patients with TNBC, demonstrated a significantly higher rates of pCR (24% vs 8% $p > 0.001$) among TNBC patients compared with the hormone receptor-positive group. Similar to that shown by Liedtke and colleagues (2008), patients who achieved a pCR also experienced improved PFS and OS (Guarneri et al., 2006).

Selection of chemotherapy is based on the traditional parameters used for breast cancer; since there are currently no evidence-based preferred regimes for TNBC. Chemotherapy has obviously been the plinth of systemic treatment for TNBC, since other modalities of endocrine and targeted therapy are not applicable. Despite the high responsiveness of TNBC to Anthracycline-based neo-adjuvant chemotherapy, there is a high risk of recurrence if residual disease remains; which accounts for the poorer outcomes in these patients.

The breast cancer susceptibility gene BRCA1 is implicated in repair of double stranded DNA breaks. The high association between TNBC and BRCA1 mutation may also be indicative of a potential sensitivity to agents which cause DNA damage (Rottenberg et al., 2006; Byrski et al. 2009). Silver et al. (2010) tested the efficacy of the DNA cross-linking agent, cisplatin in a TNBC population not enriched for BRCA-mutation carriers. Eighteen of the 28 patients had a clinical complete or partial response to therapy with 6 achieving complete pathologic remission. Two of the 6 patients who attained pCR were germline BRCA1-mutation carriers. Sirohi et al. (2008) reported a clinical response rate of 88% in TNBC after neo-adjuvant treatment with platinum-containing cytotoxic agents, compared to 55% clinical complete response rate in other breast tumours. However, the overall five-year survival was still worse for TNBC compared to tumours of other subtypes. Other studies investigating the activity of platinum agents in the neo-adjuvant therapy of TNBC is currently being studied.

Table 3. Rationale for Therapeutic Options in TNBC

Proven Agents	Therapeutic Basis
Anthracyclines	High proliferation index
Taxanes	p53 mutation
Potential Agents	
Platinum	Genetic Instability
Bevacizumab	Microvascular Proliferation
Cetuximab	EGFR Over-expression
Olaparib	Genetic Instability
Sunitinib	Tyrosine kinase expression

Targeted Therapy

Epidermal Growth Factor Receptor (EGFR) receptor is a potent stimulating factor of cell-growth-activating pathways and thus stimulates tumour growth when activated (Burgess 2008). EGFR is expressed in approximately 60% of TNBC (Siziopikou et al., 2006). There are a number of studies linking TNBC to EGFR expression, with percentages ranging from 42 to 71% of women with TNBBC having EGFR expression (Cheang et al., 2008; Meche et al., 2009; Collins et al. 2009). EGFR expression in breast cancer is associated with poor disease outcome.

Viale et al. (2009) reported worse disease-free survival (DFS), overall survival (OS) and distant disease-free survival (DDFS) for patients with TNBC having EGFR expression compared to those with tumours without EGFR expression. The significant correlation between EGFR immunoreactivity with worse prognosis in patients with triple-negative invasive ductal carcinomas supports further studies on the correlation between the degree of EGFR expression and outcome of triple negative breast (Viale et al., 2009).

The EGFR over-expression that is evident in most triple negative cancers has provided a rationale for trials of the anti-EGFR monoclonal antibody cetuximab; usually in combination with platinum (Carey et al., 2008). Most recently, the phase II BALI-1 trial conducted in six countries randomized 173 patients with metastatic TNBC who were randomized to receive cisplatin in combination with cetuximab or cisplatin alone. There was a modest yet statistically significant improvement in PFS among patients who received combination therapy, 1.5 vs 3.7 months and a doubling of the overall response rate in the combination arm (10.3% vs 20%). However, the study failed to meet its primary endpoint of greater than a 20% response among patients who received both cisplatin and cetuximab (Baselga et al., 2010). This highlights the need for further studies to examine the efficacy of single-agent platinum therapy to treat TNBC as well as the use of targeted therapies, like cetuximab, in an unselected population. Two Phase II studies are currently open to test the efficacy of cetuximab in combination with pre-operative chemotherapy, ixabepilone (NCT01097642) and docetaxel (NCT00600249). Another approach under investigation is the use of small molecules such as erlotinib

which inhibit the tyrosine kinase domain of EGFR. Erlotinib is currently being evaluated in combination with docetaxel and carboplatin in patients with metastatic TNBC (NCT00491816).

Angiogenesis is required for tumor growth, invasion and metastasis in several malignancies, including breast cancer. The vascular endothelial growth factor (VEGF) is a key mediator of angiogenesis and has been shown to be a valid target for monoclonal antibody therapy in several solid tumors. The demonstration of focal endothelial tufts on histological examination of TNBC points to the potential for the use of bevacuzimab (a humanized anti VEGF monoclonal antibody) in these tumours. In the E2100 study that evaluated this agent along with paclitaxel, in TNBC patients, the PFS was increased to 10.2 months compared to 4.7 months in the paclitaxel alone arm (O'Shaughnessy et al., 2009). Subset analyses of the AVADO and RIBBON-1 trials indicate improvement in progression free survival when bevacuzimab was combined with chemotherapy in the treatment of TNBC (Robert et al., 2009; Chan et al., 2010). Currently there are multiple Phase II/III trials designed to test the efficacy of bevacizumab in combination with Taxane or Platinum based chemotherapy in the neoadjuvant and adjuvant setting.

The enzyme poly-adenosine diphosphate ribose polymerase (PARP) is known to be involved in base-excision repair after DNA damage. The association of BRCA1 and triple negative status may be potentially exploited with therapeutic benefit by the combination of PARP inhibitors and chemotherapy. There are early indications of improvement in survival in TNBC after use of PARP inhibitors combined with gemcitabine and carboplatin. In a Phase II study by O'Shaughnessy et al. (2009) randomly were assigned patients with metastatic triple-negative breast cancer to receive carboplatin and gemcitabine alone or in combination with iniparib, an intravenous PARP inhibitor. The addition of iniparib to gemcitabine and carboplatin improved the rate of clinical benefit from 32% to 52% ($p = 0.02$), median PFS progression-free survival from 3.6 months to 5.9 months and median OS (12.3 vs 7.7 months, HR = 0.57, $p = 0.01$). among those individuals who were treated with iniparib and chemotherapy when compared to chemotherapy alone (O'Shaughnessy et al., 2011). This work launched a randomized phase III trial evaluating iniparib in combination with carboplatin and gemcitabine vs. chemotherapy alone.

Conclusion

Triple negative breast cancers lack ER, PR and do not over-express the HER2. They account for about 15% of all invasive breast cancers but are over represented blacks and Hispanics. The pathology, behavior and pattern of metastases distinguish these tumours from their receptor-positive counterparts. The inferior outcome of this disease is paradoxical in the face of the chemosensitivity of the primary tumour. The development of newer biologic and targeted therapies, such as antiangiogenic agents, EGFR inhibitors, and PARP inhibitors, continues to be a promising area of research. The sensitivity to platinum agents and the utility of newer targeted therapy directed against other receptors in both the neoadjuvant and adjuvant setting is the subject of current research.

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