Familial Breast Cancer – Targeted Therapy in Secondary and Tertiary Prevention

Karin Kast^a Kerstin Rhiem^b

^aDepartment of Gynecology and Obstetrics, University Hospital Carl Gustav Carus, Technische Universität Dresden, Germany;

^bCenter of Familial Breast and Ovarian Cancer, Department of Gynecology and Obstetrics, University Hospital Cologne, Germany

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Summary

The introduction of an increasing number of individualized molecular targeted therapies into clinical routine mirrors their importance in modern cancer prevention and treatment. Well-known examples for targeted agents are the monoclonal antibody trastuzumab and the selective estrogen receptor modulator tamoxifen. The identification of an unaltered gene in tumor tissue in colon cancer (*KRAS*) is a predictor for the patient's response to targeted therapy with a monoclonal antibody (cetuximab). Targeted therapy for hereditary breast and ovarian cancer has become a reality with the approval of olaparib for platin-sensitive late relapsed *BRCA*-associated ovarian cancer in December 2014. This manuscript reviews the status quo of poly-ADP-ribose polymerase inhibitors (PARPi) in the therapy of breast and ovarian cancer as well as the struggle for carboplatin as a potential standard of care for triple-negative and, in particular, *BRCA*-associated breast cancer. Details of the mechanism of action with information on tumor development are provided, and an outlook for further relevant research is given. The efficacy of agents against molecular targets together with the identification of an increasing number of cancer-associated genes will open the floodgates to a new era of treatment decision-making based on molecular tumor profiles. Current clinical trials involving patients with *BRCA*-associated cancer explore the efficacy of the molecular targeted therapeutics platinum and PARPi.

Introduction

The identification of biomarkers with prognostic and predictive value enables oncologists to select a more efficient and less toxic therapy for their patients on the basis of individual tumor characteristics. Data from recent clinical trials point towards 2 agents for the targeted treatment of *BRCA* mutation carriers with breast or ovarian cancer: platinum-containing chemotherapies and poly-ADP-ribose polymerase inhibitors (PARPi). In vitro both substances lead to apoptotic cell death of *BRCA*-deficient tumor cells albeit using different mechanisms. The shared drug target is the absence of homologous recombination in *BRCA*-deficient tumor cells. Homologous recombination is an error-free repair mechanism of DNA double-strand breaks (DSB) [1]. The absence of homologous recombination activates error-prone DSB mechanisms like non-homologous end joining resulting in genomic instability of the cells. Platinum compounds cause DNA crosslinks that lead to DSB. At the same time PARPi prevent single-strand break repair which is also followed by DSB [2].

The Role of Platinum-Based Chemotherapy in Patients with *BRCA*-Associated Breast Cancer

Carboplatin acts on the Achilles heel of *BRCA*-deficient tumors; they are no longer capable of homologous repair which is the most reliable DNA repair mechanism in the presence of DSB caused by platinum adducts [3–5]. Whereas healthy body cells are heterozygous for the *BRCA* germline mutation, in tumor cells due to a second hit the intact allele is lost and tumor cells are predominantly prone to apoptosis after treatment with carboplatin. Although there is not yet enough data from randomized controlled clinical trials to support platinum as standard treatment in *BRCA*-associated breast cancer, in vitro and in vivo data indicate a particular sensitivity to platinum-based therapy. In *BRCA1* carriers with

breast cancer an amazing tumor response rate after neoadjuvant chemotherapy with cisplatin was reported [6–8].

Additionally a higher response to neoadjuvant chemotherapy with cisplatin was described in triple-negative breast cancers (TNBC) with germline or somatic *BRCA1/2* mutations compared to non-cisplatin chemotherapy [9]. The reason for the increased sensitivity of TNBC to platinum might be that about 15% of these heterogenous tumors are *BRCA*-associated with mostly *BRCA1* and rarely *BRCA2* harboring the underlying mutation [10, 11]. The histopathologic features of TNBC serve as surrogate marker for high genomic instability and response to DNA-damaging agents such as the DNA crosslinkers carboplatin and cisplatin [12].

Most recently GeparSixto, a prospective randomized controlled phase II clinical trial, reported that the addition of carboplatin (weekly carboplatin, area under the curve 2) to neoadjuvant chemotherapy significantly improved the pathologic complete response (pCR) rate in patients with TNBC (n = 315) from 44 to 64% irrespective of *BRCA* status and family history [15].

Furthermore the addition of carboplatin to standard neoadjuvant chemotherapy increased pCR rates in patients with TNBC in the CALGB 40603 study [13]. In this phase II trial patients with TNBC (n = 433) received paclitaxel with or without bevacizumab and/or carboplatin. The 4 treatment arms were followed by dose-dense chemotherapy with doxorubicin and cyclophosphamide. A pCR rate of 54% was reported in patients receiving carboplatin and 41% in patients treated without carboplatin.

In addition Tutt et al. [14] at the San Antonio Breast Cancer Symposium 2014 presented the TNT trial, a phase III study in first-line treatment of patients with TNBC containing a subgroup of 43 *BRCA1* or *BRCA2* mutation carriers. After 6 cycles of carboplatin or docetaxel a longer progression-free survival (PFS) in carriers (6.8 months) compared to non-carriers (3.1 months) was demonstrated. In contrast to the data of von Minckwitz et al. [15] the TNT trial did not find a superior response with carboplatin compared to standard therapy in the whole group of TNBC patients. The reason might have been the different treatment settings comparing palliative to adjuvant therapy. Due to the intratumoral heterogeneity that derives from genomic instability and selection pressure under chemotherapy, the tumor might change its main features [11]. Even reconstitution of homologous repair in platinum-resistant ovarian cancer cells was described [16]. Advanced *BRCA*-associated breast cancer might therefore behave differently to primary early breast cancer.

Platinum-Based Chemotherapy in Patients with *BRCA*-Associated Ovarian Cancer

Moreover there is clinical evidence for the efficacy of platinum in patients with *BRCA* mutations derived from ovarian cancer trials. A pooled analysis of 26 observational studies on the survival of women with epithelial ovarian cancer (EOC) included data from 1,213 EOC cases with pathogenic germline mutations in *BRCA1* (n = 909) or *BRCA2* (n = 304) and from 2,666 non-carriers recruited

and followed up at variable times between 1987 and 2010 [17]. Among patients with invasive EOC having a germline mutation in *BRCA1* or *BRCA2* was associated with improved 5-year overall survival (OS). *BRCA2* carriers had the best prognosis. Irrespective of family history around 15% of non-mucinous ovarian carcinomas and 11–22% of high-grade serous ovarian cancers are *BRCA1/2*-associated [18, 19]. In the presence of a family history of breast or ovarian cancer mutation frequency rises to 40% and higher dependent on age of onset and number of affected relatives [20].

Platinum-Based Chemotherapy So Far No Standard in *BRCA*-Associated Breast Cancer

The above data lead to the presently observable tendency of oncologists to add platinum to chemotherapy regimens for *BRCA* mutation carriers with breast cancer outside of trial concepts, and this tendency will also increase for women with TNBC; therefore a prospectively planned randomized controlled trial is highly necessary. By means of a translational research program this trial will provide the rationale for further studies in sporadic breast cancers with a *BRCA*ness phenotype that may account for up to 20% of all breast cancers [21]. These women may also benefit from the addition of platinum compounds and other agents targeting the *BRCA* signaling pathway (e.g. PARPi). Therefore the German Consortium of Hereditary Breast and Ovarian Cancer (GC-HBOC) established the NeoFam trial (supported by the Deutsche Forschungsgemeinschaft) for comparison of weekly carboplatin with paclitaxel after standard anthracycline-containing neoadjuvant therapy of patients with *BRCA1/2*-positive early breast cancer (EudraCT number: 2014–004737–51).

The Role of PARP Inhibitors in Patients with *BRCA*-Associated Breast and Ovarian Cancer

PARPi selectively produce cell death in *BRCA*-deficient tumor cells via ‘synthetic lethality’. This term describes the inactivation of 1 of the 2 most important alternative cell mechanisms in a cancer cell which prevent fatal cell damage in the first place. The additional inactivation of the residual ‘rescuing’ mechanism by a targeted drug finally induces tumor cell death. However further models try to explain the function of PARPi including not only the involvement in single-strand break repair but also the activation of another alternative DNA repair process, non-homologous end joining. Therefore PARPi are very promising drugs within treatment concepts against *BRCA*-associated cancers and may also be efficient in cancers associated with other mutated homologous recombination genes. 3 members of the 18-member PARP family (PARP1, 2, and 3) that have been identified in mammalian cells are linked to DNA repair [22]. Most preclinical and clinical data focusses on the role of PARP1 in DNA repair, regulation of genomic stability in the cell, or involvement in cellular energy mechanisms as a target of PARPi [23]. The different models describing the cel-

lular functions of PARP and the mechanisms of PARPi to selectively kill homologous recombination-deficient tumor cells might explain why some cancers respond to PARPi and others do not. Therefore further preclinical investigations and clinical trials are needed to analyze the different antitumoral effects of PARPi.

Approval of PARPi Olaparib for *BRCA*-Associated Ovarian Cancer

The PARPi which is currently developed furthest for clinical use is olaparib (Lynparza™, AstraZeneca, Wilmington, DE, USA). It is directed against PARP1, PARP2, and PARP3. In December 2014 the FDA and EMA granted accelerated approval for maintenance therapy after platinum chemotherapy for relapsed high-grade serous ovarian/fallopian/peritoneal cancer (HGSOC) in patients with a germline (FDA, EMA) or somatic (EMA) mutation in the breast cancer genes *BRCA1* or *BRCA2*. Proof-of-concept phase I/II trials in *BRCA1/2* mutation carriers with advanced breast and ovarian cancer extended first findings about the clinical effect of single agent activity of olaparib from phase I dose escalation trials [24, 25]. Recent data from a randomized, placebo-controlled, phase II trial in a maintenance setting with patients who are most likely to benefit from PARPi due to a *BRCA* mutation and platinum-responsive relapsed ovarian cancer lead to the approval in the US and the EU. The authors observed an increased median PFS of 8.4 versus 4.8 months after 2 or more lines of platinum-based therapy. This is the first new agent that brought such an improvement in ovarian cancer since bevacizumab in 2011 [26–28]. An interim analysis with 58% maturity showed differences between olaparib and placebo, in the *BRCA1/2* mutation carriers with a hazard ratio (HR) of 0.18 (95% confidence interval 0.11–0.31) and a median PFS of 11.2 vs. 4.3 months, respectively. OS did not show a difference in this group, (HR = 0.74; median OS 34.9 vs. 31.9 months) probably due to the 22.6% of patients on placebo who switched to olaparib. Olaparib is an oral PARPi that is currently under further investigation e.g. within the SOLO1 and SOLO2 phase III trials. It is given after a platinum-containing chemotherapy. To be eligible SOLO1 patients have to display a good response to the first platinum-based chemotherapy for advanced (FIGO stage III–IV) primary ovarian, fallopian, or peritoneal cancer. The SOLO2 trial focuses on patients with platinum-sensitive relapse. At the same time accumulating data exists for prolongation of disease-free survival in HGSOC with and without mutations in *BRCA1* or *BRCA2* [29].

Two proof-of-principle trials with *BRCA* germline mutation carriers demonstrated similar response rates with olaparib in breast and ovarian cancer [24, 25]. In breast cancer current trials concentrate on palliative therapy of metastasized germline *BRCA1/2*-mutated breast cancer after several lines of chemotherapy. (Neo-)adjuvant trial concepts focus on maintenance therapy post chemotherapy and surgery (OlympiA trial and Brightness by German Breast Group). In the OlympiA trial patients with TNBC and elevated risk for recurrence receive treatment with olaparib versus placebo for 12 months after neoadjuvant chemotherapy and surgery or following

adjuvant chemotherapy. Several other trials for patients with breast cancer in different therapy indications and with a variety of PARPi, e.g. veliparib, rucaparib, niraparib, are under way. Iniparib, originally assumed to be an active PARPi, in a phase III trial in combination with carboplatin/gemcitabine in patients with metastasized breast cancer failed and is no longer considered a PARPi [30–32].

Side effects of the different effective oral PARPi are consistent with mostly grade 2 toxicity for e.g. nausea, fatigue, anemia, diarrhea, dysgeusia, and thrombocytopenia. Rare side effects include myelodysplastic syndrome, acute myeloid leukemia (AML), and pneumonitis. Because of their seriousness these side effects could impede the development of PARPi in primary prevention [33]. Future development now aims to increase bioavailability for less tablet intake per day, which ranges at present between 2×8 and 1×1 tablets.

Combined Therapy with PARPi and Carboplatin

Current therapeutic concepts of multiple PARPi focus not only on PARPi as single agents but also in combination with various DNA-damaging agents. Optimal timing of therapy and selection of patients with highest benefit beyond *BRCA* mutation carriers is still the subject of research. Studies have shown clinical benefit and interactive adverse events, including bone marrow toxicity and fatigue [29, 34, 35]. Moreover PARPi might function as a sensitizer to platinum-based chemotherapy or radiation [36]. Therefore intermittent intake of oral PARPi starting a few days before platinum-containing chemotherapy is a very interesting approach. As a next step another phase III trial, PAOLA1, is investigating the concurrent use of olaparib versus placebo with first-line platinum-containing chemotherapy plus bevacizumab in advanced high-grade ovarian/fallopian/peritoneal cancer independent of a germline mutation. In breast cancer BROCADE3 offers treatment with carboplatin and paclitaxel in combination with intermittent application of veliparib versus placebo to *BRCA1/2*-associated advanced or metastasized disease.

BRCAness: Treatment Options Beyond the Germline Mutation Status

As mentioned before PARPi might be efficient in carcinomas with impaired repair mechanism of homologous recombination. Recent data indicate that up to 50% of HGSOC might be caused by homologous repair deficiency (HRD) [37]. HRD might be the result of germline *BRCA1/2* mutations which are found in approximately 15% of EOC, somatic *BRCA1/2* mutations (approximately 7% of HGSOC), mutations in other genes affecting proteins involved in homologous recombination (e.g. *RAD51C*, *RAD51D*, *ATM*, *CHEK2*), and functional silencing of genes concerning the homologous recombination mechanism (10% of HGSOC) [11, 19–23]. This phenomenon is referred to as ‘BRCAness’, and different strategies are being followed to establish a routine test for its

detection in tumor tissue in paraffin material. So far preparation of tumor DNA and sequencing of *BRCA1/2* and other genes involved in homologous recombination is the most reliable but also the most costly strategy. Moreover mutations of unknown significance cause difficulties in the interpretation of the analysis. Great efforts are currently being made to establish a functional test for HRD. All current trials are accompanied by large biomarker projects that require collection of blood and tumor material.

Similar to HER2-directed therapies indication for treatment with PARPi might be dependent on a test which is performed on tumor tissue in paraffin. If testing of tumor material is performed first, genetic counselling is recommended for patients with somatic *BRCA1/2* mutations because a germline mutation will be found in more than half of them. A germline mutation in *BRCA1* or *BRCA2* implies the known risk for secondary cancer and cancer risk for consanguineous relatives [16].

Outlook

The inclusion of targeted agents such as platinum and PARPi in anticancer therapy of *BRCA1/2* mutation carriers and BRCAness cancers has unleashed future challenges. There are a lot of unresolved questions: How can we select the patients who are most likely to benefit? A deleterious germline mutation in the *BRCA* genes is a predictive marker for the use of PARPi. Analysis currently involves various sequencing methods and screening for large deletions or insertions by multiplex ligand probe analysis; however are there other tools with which to identify susceptible patients with methylation of *BRCA* genes or somatic mutations? Moreover

BRCA-like gene expression profiles in *BRCA1/2*-negative familial and sporadic carcinomas (e.g. TNBC) may show the same response rates to platinum or PARPi as carcinomas of *BRCA* mutation carriers. Does the impairment of other homologous recombination genes indicate potential drug response? What is the optimal timing, dosage, scheduling, and sequencing of PARPi? Could severe adverse events like AML preclude the use of PARPi in primary prevention? What are the mechanisms of resistance to PARPi, and how can they be overcome?

Conclusion

Approval of PARPi in *BRCA1/2*-associated ovarian cancer gives way to a new kind of medication that targets not only the germline mutation but also the resulting deficiency, HRD, which is often found in HGSOC and TNBC. Highly interesting trials with different oral PARPi are ongoing for both tumor entities in various therapy settings. Current trials with targeted agents are supported by concepts of identifying and validating predictive biomarkers for the stratification of patients. These data will further advance the field of targeted therapy. Besides, chemotherapy with carboplatin is becoming more and more important for the treatment of TNBC with or without *BRCA1* and *BRCA2* mutations; however further studies are needed.

Disclosure Statement

The authors have no conflicts of interest to declare.

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