BRCA Gene Mutations and Poly(ADP-Ribose) Polymerase Inhibitors in Triple-Negative Breast Cancer

13

Hitomi Sumiyoshi Okuma and Kan Yonemori

Abstract

Breast cancer is the most common cancer in women worldwide. Treatment is chosen according to its hormone receptor status and human epidermal growth factor receptor 2 (HER2) status. Among the four main clinically set subtypes, hormone receptor-negative/HER2-negative subtype, also called triple-negative subtype (TNBC), is the most aggressive type with limited choices of therapy. However, recent research has provided important new insights into effective treatments for this subtype. One molecular target that has gained attention is the BRCA gene. BRCA proteins are involved in the maintenance of genomic integrity, therefore playing an important role as a "caretaker" DNA repair protein. Approximately 5% of all breast cancer patients are BRCA mutation carriers, and among the patients with BRCA mutations, 57.1% have the clinical TNBC subtype, showing a high association between BRCA mutations and TNBCs. When cells lack either BRCA1 or BRCA2, all types of homology-directed repairs are compromised, and poly(ADP-ribose) (PAR) polymerase (PARP) acts as a backup system to maintain the genome, consequently making the cells highly sensitive to PARP1 inhibitors. PARP inhibitors have shown promising activity in preclinical and early clinical trials, and today, phase III trials are ongoing. In this chapter, we discuss the mechanism and the role of PARP inhibitors in BRCA-mutated breast cancers and further elaborate the clinical potential of PARP inhibitors as well as their barriers.

H.S. Okuma • K. Yonemori (✉)
Department of Breast and Medical Oncology,
National Cancer Center Hospital,
5-1-1 Tsukiji, Chuo-ku, Tokyo, Japan
e-mail: kyonemor@ncc.go.jp

© Springer Nature Singapore Pte Ltd. 2017
E. Song, H. Hu (eds.), *Translational Research in Breast Cancer*, Advances in Experimental Medicine and Biology 1026, https://doi.org/10.1007/978-981-10-6020-5_13

Keywords

Triple-negative breast cancer • BRCA mutation • PARP inhibitor • Synthetic lethality

13.1 Introduction

Recent researches have provided new insights into the effective treatments for breast cancer, which is the most common cancer in women worldwide. Clinically treated according to its subtype, breast cancer has four subtypes identified as follows: (1) hormone receptor positive/human epidermal growth factor receptor 2 (HER2) negative, (2) hormone receptor positive/HER2 positive, (3) hormone receptor negative/HER2 negative, and (4) hormone receptor negative/HER2 negative. The last subtype is also called triple-negative breast cancer (TNBC), one of the most aggressive types of breast cancer. Unlike hormone receptor-positive (luminal-like) subtypes, there are no targeted therapies available for patients with TNBC, which shows aggressive behaviors. Therefore, many researchers are investigating the molecular background of TNBCs, with a particular focus on *BRCA1/BRCA2* mutations.

In this chapter, we will discuss TNBC and the effects of *BRCA* mutations in this type of cancer. The roles of poly(ADP-ribose) polymerase (PARP) inhibitors in breast cancer treatment will also be elucidated.

13.2 TNBC

TNBC is defined based on immunohistochemical staining criteria. In the clinical setting, TNBC is defined to be estrogen receptor (ER) negative, progesterone receptor (PgR) negative, and human epidermal growth factor receptor 2 (HER2) negative. However, TNBC remains a heterogeneous disease that includes several intrinsic subtypes. Moreover, TNBC is known for its highly aggressive behavior and poor prognosis compared with other breast cancer subtypes [1], such as ER-positive, PgR-positive, and/or HER2-positive diseases.

13.2.1 Molecular Biological Features of TNBC

TNBC accounts for approximately 15% of all breast cancers. Compared with other subtypes, TNBC tends to occur in younger patients and exhibit large tumor burden, high nuclear grade, low BCL-2 expression, and high p53 and/or Ki-67 expression.

In 2000, Perou et al. performed a complementary DNA microarray gene profiling analysis in breast cancer and identified different molecular patterns, called “molecular portraits,” among breast cancers [2]. In this analysis, they classified breast cancers into five different intrinsic subtypes: luminal A, luminal B, HER2-enriched, basal-like, and normal. Seventy-five percent of clinically proven TNBC can be classified into the basal-like subtype. In a later publication, researchers confirmed that among TNBCs, 80% were the basal-like subtype, 3% were the luminal subtype, and 9% were the HER2-enriched subtype [3].

Among basal-like subtypes, molecules such as cytokeratin 5/6, vimentin, and laminin have been shown to be highly expressed, whereas Bcl-2 has been shown to exhibit low expression [4]. Moreover, loss of phosphatase and tensin homolog (PTEN) and the disappearance of phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha (PIK3CA) expression, retinoblastoma (RB) 1 mutations, or KRAS mutations are commonly observed in basal-like TNBC [5, 6].

Mutations or deletions in the *BRCA* gene (*BRCA1* and *BRCA2*) are also found in TNBCs.

Among basal-like subtypes, 75% have been reported to be BRCA1-associated breast cancers [7], whereas 19.5% of all TNBCs show BRCA germ line mutations [8].

13.2.2 Optimal Strategies for Treatment with Currently Approved Agents

Despite the findings of molecular subtypes among TNBC, no predictive values of the molecular subtypes have been established. Treatment is therefore selected from currently recommended agents that are approved in general breast cancer population.

Anthracyclines and taxanes remain the primary therapeutic approaches for TNBC, although there is limited evidence of success in patients treated with anthracycline- and/or taxane-containing regimens in the perioperative setting [9]. Patients who show primary or acquired resistance to key drugs may be given further chemotherapeutic agents that are not crossresistant, such as capecitabine, eribulin, gemcitabine, or vinorelbine [10–12]. The use of multidrug regimens in patients with metastatic cancer is controversial, and guidelines, such as those issued by European Breast Cancer Conference [13], recommend sequential monotherapy for advanced breast cancer. In cases where the aggressive nature of the disease calls for the need to stabilize the symptoms and reduce the risk of inner organ dysfunction, which is often noted in patients with TNBC, a multidrug regimen may be recommended rather than a single-drug regimen. Other agents that are sometimes used in TNBC therapy include platinum-based regimens [14–16] and PARP inhibitors (which are being investigated). The use of these agents has been supported by the strong association of TNBC with germ line *BRCA1* mutations.

Nonetheless, TNBC shows an aggressive behavior and very poor prognosis with limited treatment options. A biomarker-based understanding of molecular targets is required to facilitate further improvements in treatment strategies for TNBC.

13.3 BRCA Mutations

13.3.1 Functions and Mechanisms of BRCA

BRCA was first discovered in the 1990s and has been one of the most notorious and well-known cancer-related genes identified to date. It was originally considered as a tumor-suppressor gene [17]. However, further evidence shows that BRCA proteins are involved in the maintenance of genomic integrity. Therefore, instead of functioning as "gatekeeper" proteins of tumor suppressor, the BRCA family of proteins acts as "caretaker" proteins of DNA repair. Moreover, BRCA proteins are known to function in concert with other proteins, such as RAD50/Mre11 and RAD51, which play important roles in repairing DNA breaks caused by ionizing radiation [18].

During DNA replication, DNA molecules are particularly vulnerable to breakage in the single-stranded molecule portions that have not yet undergone replication near the replication fork. When an accidental breakage of the still unreplicated single-stranded DNA occurs at the replication forks, the resulting breaks are functionally equivalent to double-stranded breaks occurring in an already formed double helix. These double-stranded breaks are usually fixed by homology-directed repair (HDR). At sites of stalled replication forks where double-stranded breaks are observed, BRCA1 is located along with proliferating cell nuclear antigen (PCNA) and other DNA repair proteins, including RAD50 and RAD51 [19]. BRCA2 protein is also found at the same location, providing evidence of its collaboration in the DNA repair process [20]. When cells lack either BRCA1 or BRCA2, all types of HDR are compromised.

In mice, genetic disruption of BRCA1 function causes death during early embryogenesis, whereas mutant germ line alleles of *BRCA2* cause only partial loss of function, which results in susceptibility to lymphoid malignancies and unusual chromosomal aberrations [18]. In humans, mutant germ line alleles of either *BRCA1* or *BRCA2* lead to a natural susceptibility to breast and ovarian carcinomas [21]. In ovarian

cancer, an estimated 70–80% of cases are caused by *BRCA* mutations. Some somatic mutations in *BRCA2* are associated with prostate and colon carcinomas. Additionally, female cells lacking BRCA1 function cannot properly inactivate one of the two X chromosomes. The mechanism of X-inactivation is essential in cells of early female embryos and must persist in all linear descendants. How this loss of BRCA function intersects with its DNA repair functions and how *BRCA1* mutation inclines to generate cancer primarily in women remain unknown.

13.3.2 BRCA Mutations in TNBCs

According to an analysis published by the International Breast Cancer Linkage Consortium, 0.12% of the general population carries *BRCA1* germ line mutations [22]. In patients with breast cancer, approximately 5% of patients are *BRCA* mutation carriers. According to a retrospective study, among patients with *BRCA* mutations, 57.1% have the clinical TNBC subtype [23]. Additionally, 19.5% of TNBCs have been shown to have germ line *BRCA* mutations [8]. When the population is narrowed down to those who have familial breast cancers, defined as breast cancer with a family history of one or more first- or second-degree relatives with breast cancer that does not fit the hereditary breast cancer definition, almost half of cancers are associated with germ line transmission of *BRCA1* or *BRCA2* mutations. In addition to germ line mutations, methylation of *BRCA1* is also known to be frequently found in TNBCs [24]. In all, the findings have shown that *BRCA* mutations are highly associated with TNBCs.

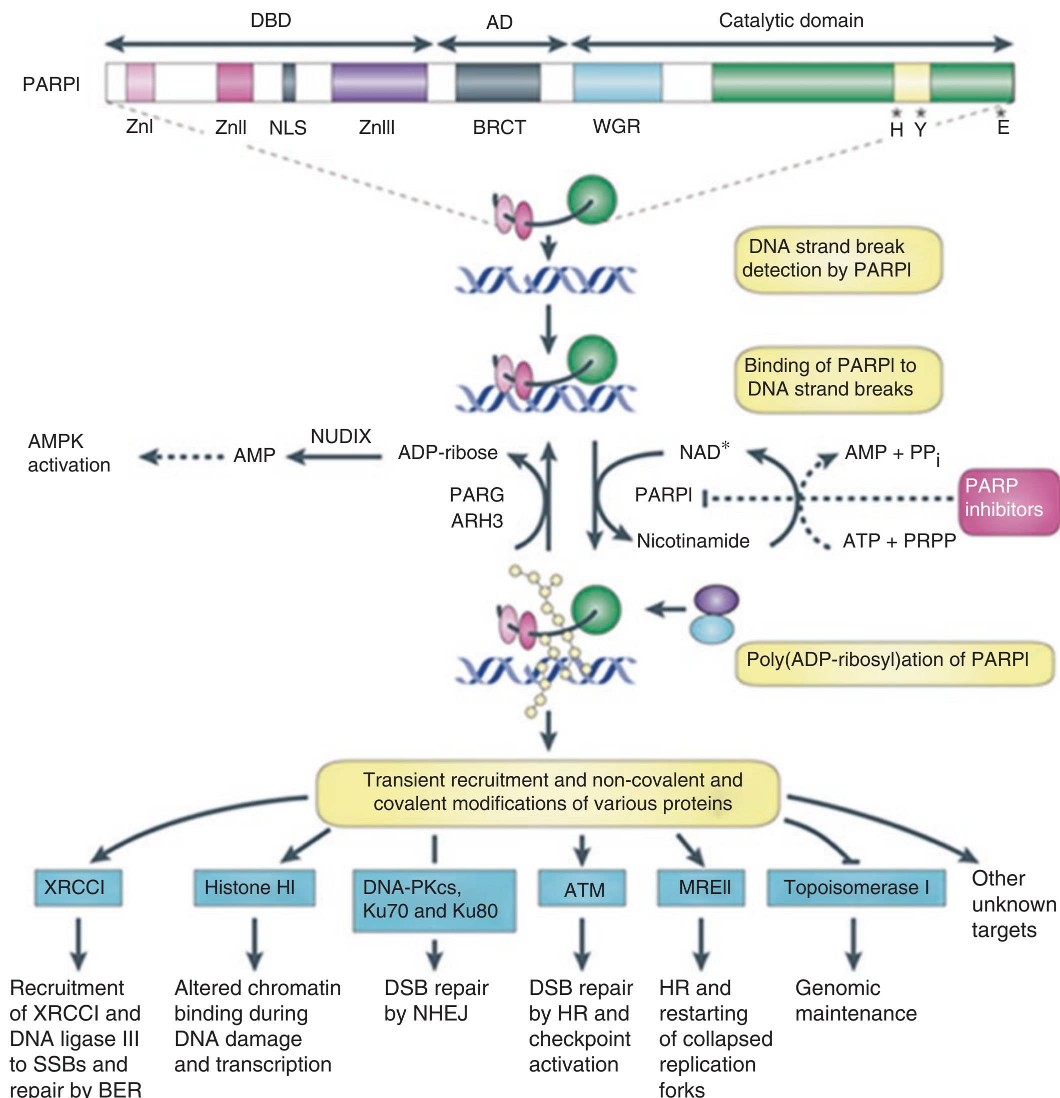
13.4 Function of PARP1

Among the many backup mechanisms required for proper repair or maintenance of the genome, poly(ADP-ribose) (PAR) synthesis is one of the earliest responses to DNA strand breakage. PARP1 is an abundant and stable component of chromatin and facilitates DNA repair by binding to DNA breaks and attracting other repairing proteins [25–29]. It is comprised of three functional domains: the amino-terminal DNA-binding domain which is important for binding of PARP1 to DNA breaks, the central automodification domain which allows the enzyme to PARate itself, and the C-terminal catalytic domain which transfers ADP-ribose subunits from NAD^+ to protein acceptors (Fig. 13.1) [30]. Among the seven main pathways used for DNA repair, PARP plays an important role in base excision repair (BER). At sites of single-stranded DNA breaks in which PARP binds to the DNA, PARP is activated and converts nicotinamide adenine dinucleotide (NAD) into ADP-ribose polymers (PAR) by attracting XRCC1, a scaffold protein that interacts with and recruits, stabilizes, or stimulates multiple enzymatic components involved in single-stranded breakage. For short patch repair and long patch repair at lesions that are more difficult to repair, the breakage goes through a single-stranded break intermediate and then arrives at a ligation stage to yield repaired DNA. PARP1 and PARP3 are among the 17 PARP isoforms that are also involved in double-stranded break repair [31].

For cells that lack BRCA1 or BRCA2 function, PARP acts as a backup system to maintain the genome and plays a critical role following accidental breaks that occur at replication forks during the S phase. Consequently, the cells become highly sensitive to killing by pharmacologic inhibitors of PARP1 [32]. However, *Parp−/−* mice are viable and fertile, which explains the redundant DNA repair systems. Therefore, PARP inhibition has little if any effect on normal tissues.

13.5 PARP Inhibitors and Their Effects on Cancer

PARP inhibitors exhibit competitive inhibition with NAD by blocking the catalytic PARP domain. PARP inhibitors show single-stranded DNA breakage repair activity, inducing apoptosis through accumulation of damaged DNA in the cells. By inhibiting PARP1, the repair



Abbreviations: *ATM*: ataxia telangiectasia-mutated, *BER*: base excision repair, *BRCT*: BRCA1 carboxy-terminal repeat motif, *DNA-PKcs*: DNA-protein kinase catalytic subunit, *DSB*: double-strand break, *HR*: homologous recombination, *NHEJ*: non-homologous end joining, *NLS*: nuclear localization signal, *PPi*: : inorganic pyrophosphate, *SSB*: single-strand break, *Zn*: zinc finger. [30]

Fig. 13.1 Function of PARP1 in DNA repair (Reprinted by permission from Macmillan Publishers Ltd: Nat Rev Cancer. (10(4): 293–301), copyright (2010)) Abbreviations: *ATM*, ataxia telangiectasia mutated; *BER*, base excision repair; *BRCT*, BRCA1 carboxy-terminal repeat motif; *DNA-PKcs*, DNA-protein kinase catalytic subunit; *DSB*, double-stranded break; *HR*, homologous recombination; *NHEJ*, nonhomologous end joining; *NLS*, nuclear localization signal; *PPi*, inorganic pyrophosphate; *SSB*, single-stranded break; *Zn*, zinc finger [30]

phenomenon can be trapped at the single-stranded intermediate state, thereby blocking ligation. PARP inhibitors bind to the catalytic site and prevent the release of PARP1 from DNA by "trapping" PARP1 at the site and removing PARP1 from the normal catalytic cycle [27, 28, 33]. When BER does not function properly, single-stranded breaks are left unrepaired, leading to

the formation of double-stranded breaks due to stalling of the replication fork. Since double-stranded breaks are repaired by either nonhomologous end joining or homologous recombination, inhibition of PARP alone does not lead to efficient cell death. Therefore, for PARP inhibitors to exert beneficial effects on DNA repair, another repair pathway other than BER must be functionally damaged by PARP inhibition.

13.5.1 Synthetic Lethality

Synthetic lethality was introduced nearly a century ago by geneticists. It involves the combined knockout of two genes, which leads to a lethal form of genetic interactions that can selectively kill cancer cells while sparing normal cells [34]. The concept of synthetic lethality involving PARP and BRCA is related to the observation that both proteins are normally nonessential but critical for the survival of cancer cells. The most striking evidence of synthetic lethality is the use of PARP inhibitors in homologous recombination-defective tumors [32, 35]. As BRCA1 and BRCA2 are associated with homologous recombination, PARP inhibitors have been used for monotherapy in treating patients with *BRCA1*- or *BRCA2*-mutated cancers. Other genes associated with homologous recombination are *RAD51*, *RAD54*, *DSS1*, *PRA1*, *NBS*, *ATR*, *ATM*, *CHK1*, *CHK2*, *FANCD2*, *FANCA*, and *FANCC*. Cells with a deficiency in one of these genes show sensitivity to PARP inhibitors, confirming the concept of synthetic lethality [33].

13.6 Clinical Application of PARP Inhibitors in Cancer

In PARP1-knockout mice, deficiencies in PARP1 function result in impaired DNA repair, which consequently leads to a higher sensitivity to anticancer agents. It indicates that PARP1 inhibition may induce sensitivity to DNA damage by anticancer agents and therefore act as a radiosensitizer or chemosensitizer in the treatment of cancers. PARP1 is also known for its strong activation by radiotherapy or DNA methylating anticancer agents. Based on available evidence, along with the development of PARP inhibitors in patients with germ line BRCA mutations, new therapeutic approaches using PARP inhibitors combined with DNA-damaging anticancer agents have been evaluated. Approximately 30 years ago, small-molecule nicotinamide analogs were found to enhance the cytotoxicity of dimethyl sulfate, a DNA-damaging agent, by inhibiting PARylation [36–38]. Subsequently, clinical PARP inhibitors, including veliparib, rucaparib, olaparib, and niraparib, were developed. A more potent second-generation PARP inhibitor, talazoparib, has also been developed [39]. The difference among these agents is the ability to “trap” PARP1, an essential mechanism of PARP inhibitors. Talazoparib is approximately 100 times more potent than niraparib and is therefore more potent than olaparib and rucaparib [40]. The chemical structures of clinical PARP inhibitors and the ability of each PARP inhibitor to “trap” PARP1 is thought to broadly correlate with its cytotoxic potency [33]. Among currently available PARP inhibitors, olaparib (Lynparza) was the first to be approved by the US Food and Drug Administration (FDA) for treating patients with germ line *BRCA* mutations in advanced ovarian cancer in February 2014. The development of olaparib in breast cancer will be further discussed in this chapter.

13.7 PARP Inhibitors in the Field of Breast Cancer

During clinical development, PARP inhibitors have been investigated in combination with DNA-damaging anticancer agents or radiotherapy, or as monotherapy, in cancers that show decreased BRCA1 or BRCA2 functions, mainly TNBC. In the field of breast cancer, BSI-201 was the first PARP1 inhibitor to be reported [41]. In a phase I (and Ib) trial, this compound showed

safety and effectiveness and was later tested in a randomized phase II trial, which compared combined treatment of gemcitabine plus carboplatin (GC) plus BSI-201 and GC alone in patients with metastatic TNBC with two or less prior regimens [42, 43]. The progression-free survival (PFS) was 6.9 months versus 3.3 months, and overall survival was 9.2 months versus 5.7 months, indicating a statistically longer survival for the GC plus BSI-201 arm. The overall response rate was also higher in the GC plus BSI-201 arm (48% versus 16%, $p = 0.002$). There were high expectations for the phase III trial, but the primary endpoint was not achieved.

Alternatively, olaparib has been developed as another promising PARP inhibitor for the treatment of breast cancer, which will be discussed below.

13.8 Development of Olaparib (Lynparza) in Breast Cancer

Olaparib is a PARP1 inhibitor first discovered during a screening test for agents that induce sensitivity of cells to cytotoxic agents, such as topoisomerase I inhibitors and alkylating agents. It has showed antitumor activity in cells with homologous recombination deficiency, which implies its role as a promising agent for the treatment of *BRCA*-mutated cancer. Moreover, olaparib was first approved by the FDA for treatment of *BRCA*-mutated ovarian cancers. In this section, we will discuss the development of olaparib studies in the field of breast cancer.

13.8.1 Preclinical Study

Through an in vitro study, olaparib monotherapy demonstrated strong antitumor activity in breast cancer cells with *BRCA1* mutations [44]. In an in vivo study of *BRCA1*−/− tumor-bearing mice, olaparib inhibited tumor growth without signs of toxicity, which significantly increased the survival rate. In a similar analysis with *BRCA2*−/− murine mammary epithelium, daily exposure to olaparib for 28 days caused significant regression or growth inhibition in 46 of 52 tumors [45]. The same analysis was conducted with olaparib in combination with carboplatin. Although no advantage over carboplatin monotherapy was observed, a significant increase in time to tumor relapse or death was observed if PARP inhibitors were continuously administered [46]. In combination therapy, temozolomide or dacarbazine plus olaparib was shown to have antitumor activity. Similarly, olaparib with topoisomerase I inhibitors or platinum agents also showed activity in vitro and in vivo.

13.8.2 Clinical Phase I Monotherapy Trials

Olaparib was first tested in early phase clinical trials for advanced solid tumors with no further standard therapy [47]. However, as the activity of this agent against *BRCA*-mutated cancers became clearer, the protocol was amended to include patients with *BRCA* mutations. Later, during the expansion phase, patients with *BRCA* mutations were specifically enrolled, and a total of 60 patients were eventually included. The dose of oral administration started at 10 mg either once daily or twice daily for 14 consecutive days in a 21-day cycle. During the higher dose phase, the drug was taken twice daily for 28 consecutive days. Dose-limiting toxicity was confirmed at doses of 400 and 600 mg twice daily. In the cohort receiving 400 mg, one patient experienced grade 3 agitation and grade 3 fatigue, and in the cohort receiving 600 mg, one patient experienced grade 4 thrombocytopenia and another patient experienced grade 3 somnolence. Overall, 21 patients with *BRCA* mutations were enrolled, and among the 19 patients with breast, ovarian, or prostate cancers, nine patients (47%) achieved a partial response, and 12 patients (63%) achieved a clinical benefit (partial response or stable disease). This was a surprisingly high response rate for a cohort that included patients with relapsed breast, ovarian,

or prostate cancer. Furthermore, patients with *BRCA* mutations did not show higher incidences of adverse events than patients having wild-type *BRCA*.

13.8.3 Clinical Phase II Monotherapy Trials

To date, three phase II trials of olaparib monotherapy have been published in the field of breast cancer. The first trial was an international collaborative trial undertaken in six countries. This trial included patients with advanced breast cancer with *BRCA1* or *BRCA2* mutations who had been given at least one prior chemotherapy regimen [48]. The study was comprised of two different dosage cohorts: 400 mg twice daily (phase I maximum tolerated dose) and 100 mg twice daily (a dose that showed activity in the phase I trial). Objective responses were observed in 11 of 27 patients (41%; 95% confidence interval [CI]: 25–59) in the first cohort and 6 of 27 patients (22%; 95% CI: 11–41) in the latter cohort. The toxicities were mainly at low grade. Therefore, these results provided positive evidence for the concept of PARP inhibition in *BRCA*-deficient breast cancers. The second trial was a multicenter trial conducted in Canada and included patients with recurrent high-grade serous or poorly differentiated ovarian carcinoma or TNBC, regardless of *BRCA1* or *BRCA2* mutation status [49]. Patients received olaparib 400 mg twice daily. Ninety-one patients were enrolled (65 with ovarian cancer and 26 with breast cancer), and among the 63 evaluable patients, objective responses were observed in 7 of 17 patients (41%; 95% CI: 22–64) with *BRCA1* or *BRCA2* mutations and 11 of 46 patients (24%; 95% CI: 14–38) without mutations. Although no objective responses were reported in patients with breast cancer, 30% of patients achieved stable disease for at least 8 weeks, with a median PFS of 54 days. The third phase II trial was an international collaborative trial that enrolled patients with germ line *BRCA1* or *BRCA2* mutations with recurrent breast, ovarian, pancreatic, or prostate cancer [50]. Patients with breast cancer had to have at least three prior-chemotherapy regimens for metastatic disease. Olaparib was administered at 400 mg twice daily. Among the 298 patients treated and evaluated, an objective response was achieved in 78 of 298 patients (26.2%; 95% CI: 21.3–31.6) and in eight of 62 patients (12.9%; 95% CI: 5.7–23.9) with breast cancer. Stable disease was observed in 47% (95% CI: 34.0–59.9) of patients with breast cancer. Table 13.1 summarizes the phase II trials that included patients with breast cancer.

Table 13.1 Clinical phase II studies of olaparib monotherapy in breast cancer

Published year	Author	Eligibility	Olaparib dose (twice daily)	N	Response rate	PFS	Notes
2010	Tutt et al.	Advanced, BRCA mutation	400 mg	27	41%	5.7 months	
			100 mg	27	22%	3.8 months	
2011	Gelmon et al.	Advanced, BRCA mutation or TNBC	400 mg	23	0%	54 days	Stable disease of over 8 weeks: 30%
2015	Kaufman et al.	Advanced, BRCA mutation	400 mg	62	13%	3.7 months	Partial response + stable disease of over 8 weeks: 60%

PFS progression-free survival

13.8.4 Clinical Phase III Monotherapy Trials

Three phase III trials of olaparib monotherapy have been initiated in patients with germ line *BRCA* mutation-positive breast cancer. They are OlympiA (NCT02032823), Neo-Olympia (D081EC00005), and OlympiAD (NCT0000622). OlympiA is a randomized double-blind study which assesses the efficacy of olaparib at a dose of 300 mg twice daily. In this study, olaparib was administered with and without placebo as adjuvant treatment in patients with *BRCA1/BRCA2* mutations and high-risk HER2-negative breast cancer. The patients were divided into two groups, with one completing definitive local treatment and the other undergoing either neoadjuvant or adjuvant chemotherapy. Neo-Olympia is a randomized three-arm trial comparing olaparib monotherapy at a dose of 300 mg twice daily, placebo therapy plus weekly paclitaxel (80 mg/m2), and olaparib therapy at a dose of 100 mg twice daily plus weekly paclitaxel (80 mg/m2) in the neoadjuvant setting in patients with *BRCA1/BRCA2* mutations and operable, locally advanced, or inflammatory breast cancer. OlympiAD is a randomized open-label trial which assesses the efficacy of olaparib at a dose of 300 mg twice daily. It compares olaparib monotherapy with treatment of physician's choice (TPC) of capecitabine, vinorelbine, or eribulin in patients with *BRCA1/BRCA2* mutations and metastatic breast cancer. Two of the trials began enrolment in 2014, and findings from the OlympiAD trial were recently reported at the 2017 ASCO Annual Meeting [51]. At 77% data maturity, PFS was significantly longer in the olaparib arm [7.0 vs 4.2 months, hazard ratio (HR) 0.58; 95% CI: 0.43–0.80; $p = 0.0009$] with a higher objective response rate of 59.9% in the olaparib arm compared to 28.8% in the TPC arm (HR 0.57; 95CI: 0.40–0.83). The safety profile of olaparib was consistent with prior studies. These promising results were the first to demonstrate improved outcomes with a PARP inhibitor in breast cancer. Table 13.2 summarizes the phase III trials that included patients with breast cancer.

13.8.5 Combination Therapy

Olaparib has been tested with several other agents, such as paclitaxel, temozolomide, dacarbazine, topotecan, bevacizumab, paclitaxel plus carboplatin, and newer agents (e.g., phosphoinositol 3-kinase [PI3K] inhibitors).

13.8.5.1 Paclitaxel Plus Olaparib

In a phase I/II trial, patients with advanced TNBC were treated with olaparib at a dose of 200 mg twice daily in combination with paclitaxel (90 mg/m^2, days 1, 8, and 15) on a 28-day cycle [52]. Patients were treated with either first-line or second-line chemotherapy. The response rate was high, with seven (37%) out of 19 patients achieving an objective response. Although the toxicities were relatively well tolerated, severe neutropenia was observed at a greater frequency than expected. In the second cohort, the dose intensity of paclitaxel was not retained, even with the use of prophylactic granulocyte colony-stimulating factor.

13.8.5.2 Paclitaxel Plus Carboplatin Plus Olaparib

In a cohort of patients with advanced solid tumors including breast cancer, a phase I study was conducted to investigate the treatment of olaparib with either paclitaxel (80 mg/m^2, days 1, 8, and 15) or carboplatin (AUC 4–5, day 1) or both paclitaxel (90–175 mg/m^2, day 1) plus carboplatin (AUC 4–5, day 1; TC). Olaparib was given at a dose of 50–200 mg twice a day every day or 200–400 mg twice a day for 5 or 10 consecutive days [53]. The hematological toxicities were too strong to maintain the dose in the cohorts taking olaparib every day plus carboplatin or taking olaparib everyday plus TC. However, olaparib given at a dose of 100 mg twice a day every day in combination with PTX was well tolerated, as was olaparib given at 200 mg twice a day for 10 consecutive days plus TC. The overall objective response rate was 16.1% (14/87 patients), whereas the response rate in patients with *BRCA1/BRCA2* mutations was 50% (6/12 patients).

Table 13.2 Clinical phase III studies of olaparib monotherapy in breast cancer

Trial	Eligibility	Setting	Olaparib monotherapy arm	Comparator arm(s)	Primary endpoint
OlympiA	High-risk after definitive local treatment, BRCA mutation	Adjuvant	300 mg twice daily	Placebo	Invasive disease-free survival
Neo-Olympia	Operable, BRCA mutation	Neoadjuvant	300 mg twice daily (arm A)	Placebo + weekly PTX (arm B)	Pathological complete response
				Olaparib 100 mg twice daily + weekly PTX (arm C)	
OlympiAD	Advanced, BRCA mutation	Metastatic	300 mg twice daily	Capecitabine or vinorelbine or eribulin (physician's choice)	PFS

PTX paclitaxel, *PFS* progression-free survival

13.8.5.3 Eribulin Plus Olaparib

Eribulin mesylate is a nontaxane inhibitor of microtubule dynamics of the halichondrin class of antitumor agents. Eribulin is currently recognized as a global standard treatment for metastatic or recurrent breast cancer following the use of anthracyclines and taxanes. Pooled analyses of two phase III trials of eribulin monotherapy in patients with metastatic or recurrent breast cancer suggested favorable survival benefits, particularly in patients with TNBC [11, 12]. In a cohort of patients with TNBC, a phase I/II trial was conducted in Japan to investigate the safety profiles and efficacy of olaparib in combination with eribulin under the assumption that this combination may be a favorable regimen for patients with metastatic or recurrent TNBC [54]. Patients who had received both anthracycline- and taxane-containing regimens were enrolled to be treated with eribulin at a dose of 1.4 mg/m^2 (days 1 and 8) plus olaparib twice daily every day at a dose of 25–300 mg. The recommended phase II dose of olaparib was 300 mg twice daily. Pharmacokinetic (PK)/pharmacodynamic (PD) analysis also showed that the C_{max} and area under the curve (AUC) of olaparib were dose dependent and that both parameters of eribulin and olaparib were not influenced by each other. An objective response was observed in seven of the 18 evaluable patients, indicating a relatively high response rate of 38.9% (95% CI: 17.3–64.3). Six patients maintained their responses for over a year, and the median PFS was 4.22 months (95% CI: 2.99–7.36). The most frequent adverse events were the occurrences of neutropenia (grade 3 or more: 83.3%), but the drug was overall well tolerated.

13.9 Development of Other PARP Inhibitors: Talazoparib

Talazoparib has a much higher potency for "trapping" PARP inhibitors than olaparib. In a recent phase I study, talazoparib has shown some promise in treating 13 early-stage patients with germ line *BRCA1* or *BRCA2* mutations. The patients were treated for 2 months with talazoparib before neoadjuvant chemotherapy and surgery [55]. Decreased tumor volume was observed in all 13 patients following the 2-month treatment with talazoparib, and the average volume reduction was 78% (range: 30–98%). The toxicity of this drug also proved to be well tolerated, as no grade 4 toxicities were observed, and only one patient required dose reduction due to grade 3 neutropenia. The study is ongoing, and researchers will next

Table 13.3 Clinical studies of PARP inhibitors including breast cancer

Drug	Phase	Eligibility	Concomitant therapy	Notes
Olaparib	I	Breast cancer or ovarian cancer	Carboplatin	BRCA1 or BRCA2 mutation
	I	Breast cancer or women's cancer	Carboplatin	
	I	TNBC or ovarian cancer	BKM120	
	I	Solid tumors, including TNBC	Carboplatin and/or PTX	
	I/II	TNBC	PTX	
	I/II	TNBC or ovarian cancer	Cediranib	
Iniparib	II	TNBC with brain lesion	Irinotecan	
	II	TNBC	Gemcitabine and carboplatin	Iniparib twice weekly versus weekly
	II	TNBC	PTX	Neoadjuvant
Veliparib	I	Solid tumors	TMZ	BRCA1- or BRCA2-mutated breast cancer
	I	TNBC or gynecologic cancer	Pegylated liposomal doxorubicin	
	I	Breast cancer	Radiation therapy	Loco-regionally recurrent
	II	Breast cancer	TMZ	BRCA1- or BRCA2-mutated breast cancer
	II	TNBC or ovarian or non-Hodgkin's lymphoma	Cyclophosphamide	
Talazoparib	I	Solid tumors		
	III	Breast cancer		BRCA1 or BRCA2 mutation (versus physician's choice)
Rucaparib	II	TNBC	Cisplatin	BRCA1 or BRCA2 mutation (versus cisplatin)
E7449	I/II	Solid tumors, including TNBC	Alone or plus TMZ or plus carboplatin and PTX	

TNBC triple-negative breast cancer, *PTX* paclitaxel, *TMZ* temozolomide

investigate the pathological response to talazoparib alone for 4–6 months.

Although talazoparib can kill *BRCA*-mutated cells in vitro at a 200-fold lower dose than olaparib or rucaparib, the in vitro therapeutic ratio achieved in *BRCA1-/BRCA2*-defective cells is similar with that in wild-type cells for all three PARP inhibitors. Therefore, it is still too early to draw any conclusion regarding which PARP inhibitor is most effective. Table 13.3 shows the clinical trials conducted with PARP inhibitors in patients with breast cancer (excluding the clinical trial of olaparib monotherapy discussed above).

13.10 Acquired Resistance to PARP Inhibitors

Multiple potential mechanisms of resistance have been identified through in vitro experiments. Even though homologous recombination repair is defective, the restoration of homologous recombination repair in *BRCA1*-mutant tumor cells has been identified through loss of 53BP1 and REV7 proteins [56, 57]. Moreover, the loss of PARP1 [58] has been proposed to cause resistance, as with other proteins that are important for maintaining replication fork stability [59]. Secondary mutations in *BRCA1* or *BRCA2* can also occur, leading to restoration of sufficient homologous recombination repair function and resulting in PARP inhibitor resistance [60, 61]. Additionally, this secondary mutation is known to cause clinical resistance to platinum-based chemotherapy [62, 63].

13.10.1 Genetic Deficiencies Other Than BRCA1/BRCA2

Not long after the discovery that *BRCA1* and *BRCA2* mutant cells were highly susceptible to PARP inhibitors, deficiencies in a number of tumor-suppressor genes, such as *ATM*, *ATR*, *PALB2*, and *FANC*, which are all involved in homologous recombination repair, have been shown to confer sensitivity to PARP inhibitors [63, 64].

In an in vitro experiment, wild-type BRCA1/BRCA2 breast cancer cells (i.e., MCF-7 and ZR-75–1 cells) that were genetically manipulated to knockdown ATM expression were treated with olaparib [65]. ATM depletion sensitized both cell lines, as assessed by short- and long-term survival assays. These data indicated that ATM depletion could sensitize breast cancer cells to PARP inhibitors and that cancers, such as those arising in mutant ATM heterozygous carriers, may be potential targets for PARP inhibitors. A similar phenomenon has been discovered for other tumor cells, such as gastric cancer cell lines and colorectal cell lines, and studies have highlighted the clinical utility of ATM expression as a predictive marker for the sensitivity of gastric cancer cells to PARP inhibitors [66].

The Fanconi anemia (FA) repair pathway is also known to play a collaborative role with *BRCA* genes. Patients with FA have a high incidence of malignancies, and their cells show hypersensitivity to DNA cross-linking agents, such as mitomycin C (MMC) and cisplatin. Cancers with defective FA/BRCA pathways are likely to be more sensitive to these types of therapy or to treatments in which an additional repair mechanism is targeted, such as treatment with PARP inhibitors. In a recent study, researchers developed a new assay to identify patients with FA functional defects using FA triple-stain immunofluorescence (FATSI, FancD2/DAPI/Ki67) [67]. The study was also conducted to verify the safety and feasibility of veliparib as monotherapy and in combination with MMC. According to FATSI screening, 28.7% (185/643) of patients were FATSI-negative, demonstrating that a substantial number of tumors exhibited FA functional deficiency. Among the 61 FATSI-negative patients who received treatment, six antitumor responses were observed with five in the combination arm. However, some clinical benefits were observed, and a better understanding of this mechanism is needed.

13.11 Concluding Remarks and Future Perspectives

Many studies have investigated the use of PARP inhibitors in breast cancer, with a particular focus on TNBC with *BRCA* mutations. So far, one trial of olaparib monotherapy has shown promising results for breast cancer. However, given the relatively small size of the study, it is difficult to tell which subset of patients would benefit the most from olaparib. Determining the optimal use of PARP inhibitors within drug combinations has been challenging, and new biomarkers may be needed to identify appropriate populations who may benefit most from PARP inhibitors. In addition, resistance to PARP inhibitors can arise in advanced disease, and further studies are needed to elucidate the related mechanisms.

References

1. Ismail-Khan R, Bui MM (2010) A review of triple-negative breast cancer. Cancer Control 17(3):173–176
2. Perou CM, Sorlie T, Eisen MB, van de Rijn M, Jeffrey SS, Rees CA, Pollack JR, Ross DT, Johnsen H, Akslen LA, Fluge O, Pergamenschikov A, Williams C, Zhu SX, Lonning PE, Borresen-Dale AL, Brown PO, Botstein D (2000) Molecular portraits of human breast tumours. Nature 406(6797):747–752. https://doi.org/10.1038/35021093
3. Cancer Genome Atlas N (2012) Comprehensive molecular portraits of human breast tumours. Nature 490(7418):61–70. https://doi.org/10.1038/nature11412
4. Han W, Jung EM, Cho J, Lee JW, Hwang KT, Yang SJ, Kang JJ, Bae JY, Jeon YK, Park IA, Nicolau M, Jeffrey SS, Noh DY (2008) DNA copy number alterations and expression of relevant genes in triple-negative breast cancer. Genes Chromosomes Cancer 47(6):490–499. https://doi.org/10.1002/gcc.20550
5. Perren A, Weng LP, Boag AH, Ziebold U, Thakore K, Dahia PL, Komminoth P, Lees JA, Mulligan LM, Mutter GL, Eng C (1999) Immunohistochemical evidence of loss of PTEN expression in primary ductal adenocarcinomas of the breast. Am J Pathol 155(4):1253–1260. https://doi.org/10.1016/S0002-9440(10)65227-3
6. Kotsopoulos J, Lubinski J, Lynch HT, Neuhausen SL, Ghadirian P, Isaacs C, Weber B, Kim-Sing C, Foulkes WD, Gershoni-Baruch R, Ainsworth P, Friedman E, Daly M, Garber JE, Karlan B, Olopade OI, Tung N, Saal HM, Eisen A, Osborne M, Olsson H, Gilchrist D, Sun P, Narod SA (2005) Age at menarche and the risk of breast cancer in BRCA1 and BRCA2 mutation carriers. Cancer Causes Control 16(6):667–674. https://doi.org/10.1007/s10552-005-1724-1
7. Foulkes WD, Brunet JS, Stefansson IM, Straume O, Chappuis PO, Begin LR, Hamel N, Goffin JR, Wong N, Trudel M, Kapusta L, Porter P, Akslen LA (2004) The prognostic implication of the basal-like (cyclin E high/p27 low/p53+/glomeruloid-microvascular-proliferation+) phenotype of BRCA1-related breast cancer. Cancer Res 64(3):830–835
8. Gonzalez-Angulo AM, Timms KM, Liu S, Chen H, Litton JK, Potter J, Lanchbury JS, Stemke-Hale K, Hennessy BT, Arun BK, Hortobagyi GN, Do KA, Mills GB, Meric-Bernstam F (2011) Incidence and outcome of BRCA mutations in unselected patients with triple receptor-negative breast cancer. Clin Cancer Res 17(5):1082–1089. https://doi.org/10.1158/1078-0432.CCR-10-2560
9. Palmieri C, Krell J, James CR, Harper-Wynne C, Misra V, Cleator S, Miles D (2010) Rechallenging with anthracyclines and taxanes in metastatic breast cancer. Nat Rev Clin Oncol 7(10):561–574. https://doi.org/10.1038/nrclinonc.2010.122
10. Jassem J, Carroll C, Ward SE, Simpson E, Hind D (2009) The clinical efficacy of cytotoxic agents in locally advanced or metastatic breast cancer patients pretreated with an anthracycline and a taxane: a systematic review. Eur J Cancer 45(16):2749–2758. https://doi.org/10.1016/j.ejca.2009.05.035
11. Kaufman PA, Awada A, Twelves C, Yelle L, Perez EA, Velikova G, Olivo MS, He Y, Dutcus CE, Cortes J (2015) Phase III open-label randomized study of eribulin mesylate versus capecitabine in patients with locally advanced or metastatic breast cancer previously treated with an anthracycline and a taxane. J Clin Oncol 33(6):594–601. https://doi.org/10.1200/JCO.2013.52.4892
12. Cortes J, O'Shaughnessy J, Loesch D, Blum JL, Vahdat LT, Petrakova K, Chollet P, Manikas A, Dieras V, Delozier T, Vladimirov V, Cardoso F, Koh H, Bougnoux P, Dutcus CE, Seegobin S, Mir D, Meneses N, Wanders J, Twelves C, investigators E (2011) Eribulin monotherapy versus treatment of physician's choice in patients with metastatic breast cancer (EMBRACE): a phase 3 open-label randomised study. Lancet 377 (9769):914–923. doi:https://doi.org/10.1016/S0140-6736(11)60070-6
13. Cardoso F, Bedard PL, Winer EP, Pagani O, Senkus-Konefka E, Fallowfield LJ, Kyriakides S, Costa A, Cufer T, Albain KS, Force E-MT (2009) International guidelines for management of metastatic breast cancer: combination vs sequential single-agent chemotherapy. J Natl Cancer Inst 101(17):1174–1181. https://doi.org/10.1093/jnci/djp235
14. Foulkes WD, Smith IE, Reis-Filho JS (2010) Triple-negative breast cancer. N Engl J Med 363(20):1938–1948. https://doi.org/10.1056/NEJMra1001389
15. Uhm JE, Park YH, Yi SY, Cho EY, Choi YL, Lee SJ, Park MJ, Lee SH, Jun HJ, Ahn JS, Kang WK, Park K, Im YH (2009) Treatment outcomes and clinicopathologic characteristics of triple-negative breast cancer patients who received platinum-containing chemotherapy. Int J Cancer 124(6):1457–1462. https://doi.org/10.1002/ijc.24090
16. Tutt A, Ellis P, Kilburn L, Gilett C, Pinder S, Abraham J, Barrett S, Barrett-Lee P, Chan S, Cheang M, Dowsett M, Fox L, Gazinska P, Grigoriadis A, Gutin A, Harper-Wynne C, Hatton M, Kernaghan S, Lanchbury J, Morden J, Owen J, Parikh J, Parker P, Rahman N, Roylance R, Shaw A, Smith I, Thompson R, Timms K, Tovey H, Wardley A, Wilson G, Harries M, Bliss J (2015) Abstract S3-01: the TNT trial: a randomized phase III trial of carboplatin (C) compared with docetaxel (D) for patients with metastatic or recurrent locally advanced triple negative or BRCA1/2 breast cancer (CRUK/07/012). Cancer Res 75(9 Suppl):S3-01-S03-01. doi:https://doi.org/10.1158/1538-7445.sabcs14-s3-01
17. Scully R, Livingston DM (2000) In search of the tumour-suppressor functions of BRCA1 and BRCA2. Nature 408(6811):429–432. https://doi.org/10.1038/35044000
18. Starita LM, Parvin JD (2003) The multiple nuclear functions of BRCA1: transcription, ubiquitination and DNA repair. Curr Opin Cell Biol 15(3):345–350

19. Gudmundsdottir K, Ashworth A (2006) The roles of BRCA1 and BRCA2 and associated proteins in the maintenance of genomic stability. Oncogene 25(43):5864–5874. https://doi.org/10.1038/sj.onc.1209874
20. Wilson JH, Elledge SJ (2002) Cancer. BRCA2 enters the fray. Science 297(5588):1822–1823. https://doi.org/10.1126/science.1077171
21. Chen S, Parmigiani G (2007) Meta-analysis of BRCA1 and BRCA2 penetrance. J Clin Oncol 25(11):1329–1333. https://doi.org/10.1200/JCO.2006.09.1066
22. Ford D, Easton DF, Peto J (1995) Estimates of the gene frequency of BRCA1 and its contribution to breast and ovarian cancer incidence. Am J Hum Genet 57(6):1457–1462
23. Atchley DP, Albarracin CT, Lopez A, Valero V, Amos CI, Gonzalez-Angulo AM, Hortobagyi GN, Arun BK (2008) Clinical and pathologic characteristics of patients with BRCA-positive and BRCA-negative breast cancer. J Clin Oncol 26(26):4282–4288. https://doi.org/10.1200/JCO.2008.16.6231
24. Birgisdottir V, Stefansson OA, Bodvarsdottir SK, Hilmarsdottir H, Jonasson JG, Eyfjord JE (2006) Epigenetic silencing and deletion of the BRCA1 gene in sporadic breast cancer. Breast Cancer Res 8(4):R38. https://doi.org/10.1186/bcr1522
25. De Vos M, Schreiber V, Dantzer F (2012) The diverse roles and clinical relevance of PARPs in DNA damage repair: current state of the art. Biochem Pharmacol 84(2):137–146. https://doi.org/10.1016/j.bcp.2012.03.018
26. Krishnakumar R, Kraus WL (2010) The PARP side of the nucleus: molecular actions, physiological outcomes, and clinical targets. Mol Cell 39(1):8–24. https://doi.org/10.1016/j.molcel.2010.06.017
27. Eustermann S, Wu WF, Langelier MF, Yang JC, Easton LE, Riccio AA, Pascal JM, Neuhaus D (2015) Structural basis of detection and signaling of DNA single-strand breaks by human PARP-1. Mol Cell 60(5):742–754. https://doi.org/10.1016/j.molcel.2015.10.032
28. Dawicki-McKenna JM, Langelier MF, DeNizio JE, Riccio AA, Cao CD, Karch KR, McCauley M, Steffen JD, Black BE, Pascal JM (2015) PARP-1 activation requires local unfolding of an autoinhibitory domain. Mol Cell 60(5):755–768. https://doi.org/10.1016/j.molcel.2015.10.013
29. Satoh MS, Lindahl T (1992) Role of poly(ADP-ribose) formation in DNA repair. Nature 356(6367):356–358. https://doi.org/10.1038/356356a0
30. Rouleau M, Patel A, Hendzel MJ, Kaufmann SH, Poirier GG (2010) PARP inhibition: PARP1 and beyond. Nat Rev Cancer 10(4):293–301. https://doi.org/10.1038/nrc2812
31. Ame JC, Spenlehauer C, de Murcia G (2004) The PARP superfamily. BioEssays 26(8):882–893. https://doi.org/10.1002/bies.20085
32. Farmer H, McCabe N, Lord CJ, Tutt AN, Johnson DA, Richardson TB, Santarosa M, Dillon KJ, Hickson I, Knights C, Martin NM, Jackson SP, Smith GC, Ashworth A (2005) Targeting the DNA repair defect in BRCA mutant cells as a therapeutic strategy. Nature 434(7035):917–921. https://doi.org/10.1038/nature03445
33. Lord CJ, Ashworth A (2017) PARP inhibitors: synthetic lethality in the clinic. Science 355(6330):1152–1158. https://doi.org/10.1126/science.aam7344
34. Helleday T, Petermann E, Lundin C, Hodgson B, Sharma RA (2008) DNA repair pathways as targets for cancer therapy. Nat Rev Cancer 8(3):193–204. https://doi.org/10.1038/nrc2342
35. Bryant HE, Schultz N, Thomas HD, Parker KM, Flower D, Lopez E, Kyle S, Meuth M, Curtin NJ, Helleday T (2005) Specific killing of BRCA2-deficient tumours with inhibitors of poly(ADP-ribose) polymerase. Nature 434(7035):913–917. https://doi.org/10.1038/nature03443
36. Shall S (1975) Seminar on poly(ADP-ribose) and ADP-ribosylation of protine. J Biochem 77(Suppl):2
37. M R Purnell WJW (1980) Novel inhibitors of poly(ADP-ribose) synthetase. Biochem J 185(3):775–777
38. Terada M, Fujiki H, Marks PA, Sugimura T (1979) Induction of erythroid differentiation of murine erythroleukemia cells by nicotinamide and related compounds. Proc Natl Acad Sci U S A 76(12):6411–6414
39. Shen Y, Rehman FL, Feng Y, Boshuizen J, Bajrami I, Elliott R, Wang B, Lord CJ, Post LE, Ashworth A (2013) BMN 673, a novel and highly potent PARP1/2 inhibitor for the treatment of human cancers with DNA repair deficiency. Clin Cancer Res 19(18):5003–5015. https://doi.org/10.1158/1078-0432.CCR-13-1391
40. Murai J, Huang SY, Renaud A, Zhang Y, Ji J, Takeda S, Morris J, Teicher B, Doroshow JH, Pommier Y (2014) Stereospecific PARP trapping by BMN 673 and comparison with olaparib and rucaparib. Mol Cancer Ther 13(2):433–443. https://doi.org/10.1158/1535-7163.MCT-13-0803
41. J.J. Mahany NL, E.I. Heath et al. (2008) A phase IB study evaluating BSI-201 in combination with chemotherapy in subjects with advanced solid tumors. J Clin Oncol 26(Suppl; abstr 3579)
42. O'Shaughnessy J, Osborne C, Pippen JE, Yoffe M, Patt D, Rocha C, Koo IC, Sherman BM, Bradley C (2011) Iniparib plus chemotherapy in metastatic triple-negative breast cancer. N Engl J Med 364(3):205–214. https://doi.org/10.1056/NEJMoa1011418
43. O'Shaughnessy J, Schwartzberg L, Danso MA, Miller KD, Rugo HS, Neubauer M, Robert N, Hellerstedt B, Saleh M, Richards P, Specht JM, Yardley DA, Carlson RW, Finn RS, Charpentier E, Garcia-Ribas I, Winer EP (2014) Phase III study of iniparib plus gemcitabine and carboplatin versus gemcitabine and carboplatin in patients with metastatic triple-negative breast cancer. J Clin Oncol 32(34):3840–3847. https://doi.org/10.1200/JCO.2014.55.2984
44. Menear KA, Adcock C, Boulter R, Cockcroft XL, Copsey L, Cranston A, Dillon KJ, Drzewiecki J, Garman S, Gomez S, Javaid H, Kerrigan F, Knights C, Lau A, Loh VM Jr, Matthews IT, Moore S, O'Connor MJ, Smith GC, Martin NM (2008) 4-[3-(4-cyclopropanecarbonylpiperazine-1-carbonyl)-4-fluorobenzyl]

-2H-phthalazin- 1-one: a novel bioavailable inhibitor of poly(ADP-ribose) polymerase-1. J Med Chem 51(20):6581–6591. https://doi.org/10.1021/jm8001263
45. Hay T, Matthews JR, Pietzka L, Lau A, Cranston A, Nygren AO, Douglas-Jones A, Smith GC, Martin NM, O'Connor M, Clarke AR (2009) Poly(ADP-ribose) polymerase-1 inhibitor treatment regresses autochthonous Brca2/p53-mutant mammary tumors in vivo and delays tumor relapse in combination with carboplatin. Cancer Res 69(9):3850–3855. https://doi.org/10.1158/0008-5472.CAN-08-2388
46. Rottenberg S, Jaspers JE, Kersbergen A, van der Burg E, Nygren AO, Zander SA, Derksen PW, de Bruin M, Zevenhoven J, Lau A, Boulter R, Cranston A, O'Connor MJ, Martin NM, Borst P, Jonkers J (2008) High sensitivity of BRCA1-deficient mammary tumors to the PARP inhibitor AZD2281 alone and in combination with platinum drugs. Proc Natl Acad Sci U S A 105(44):17079–17084. https://doi.org/10.1073/pnas.0806092105
47. Fong PC, Boss DS, Yap TA, Tutt A, Wu P, Mergui-Roelvink M, Mortimer P, Swaisland H, Lau A, O'Connor MJ, Ashworth A, Carmichael J, Kaye SB, Schellens JH, de Bono JS (2009) Inhibition of poly(ADP-ribose) polymerase in tumors from BRCA mutation carriers. N Engl J Med 361(2):123–134. https://doi.org/10.1056/NEJMoa0900212
48. Tutt A, Robson M, Garber JE, Domchek SM, Audeh MW, Weitzel JN, Friedlander M, Arun B, Loman N, Schmutzler RK, Wardley A, Mitchell G, Earl H, Wickens M, Carmichael J (2010) Oral poly(ADP-ribose) polymerase inhibitor olaparib in patients with BRCA1 or BRCA2 mutations and advanced breast cancer: a proof-of-concept trial. Lancet 376(9737):235–244. https://doi.org/10.1016/S0140-6736(10)60892-6
49. Gelmon KA, Tischkowitz M, Mackay H, Swenerton K, Robidoux A, Tonkin K, Hirte H, Huntsman D, Clemons M, Gilks B, Yerushalmi R, Macpherson E, Carmichael J, Oza A (2011) Olaparib in patients with recurrent high-grade serous or poorly differentiated ovarian carcinoma or triple-negative breast cancer: a phase 2, multicentre, open-label, non-randomised study. Lancet Oncol 12(9):852–861. https://doi.org/10.1016/S1470-2045(11)70214-5
50. Kaufman B, Shapira-Frommer R, Schmutzler RK, Audeh MW, Friedlander M, Balmana J, Mitchell G, Fried G, Stemmer SM, Hubert A, Rosengarten O, Steiner M, Loman N, Bowen K, Fielding A, Domchek SM (2015) Olaparib monotherapy in patients with advanced cancer and a germline BRCA1/2 mutation. J Clin Oncol 33(3):244–250. https://doi.org/10.1200/JCO.2014.56.2728
51. Robson ME, Im SA, Senkus E, Xu B, Domchek SM, Masuda N, Delaloge S, Li W, Tung NM, Armstrong A, Wu W, Goessl CD, Runswick S, Conte PF (2017) OlympiAD: Phase III trial of olaparib monotherapy versus chemotherapy for patients (pts) with HER2-negative metastatic breast cancer (mBC) and a germline BRCA mutation (gBRCAm). J Clin Oncol (Suppl; abstr LBA4):35
52. Dent RA, Lindeman GJ, Clemons M, Wildiers H, Chan A, McCarthy NJ, Singer CF, Lowe ES, Watkins CL, Carmichael J (2013) Phase I trial of the oral PARP inhibitor olaparib in combination with paclitaxel for first- or second-line treatment of patients with metastatic triple-negative breast cancer. Breast Cancer Res 15(5):R88. https://doi.org/10.1186/bcr3484
53. van der Noll R AJ, Jager A, et al. (2013) Phase I study of olaparib in combination with carboplatin and/or paclitaxel in patients with advanced solid tumors. J Clin Oncol 31(Suppl; abstr 2579)
54. Takahashi M YK, Yamamoto H, et al. (2016) A phase I/II trial of olaparib in combination with eribulin in patients with advanced or metastatic triple negative breast cancer (TNBC) previously treated with anthracyclines and taxanes: the analyses of efficacy and safety from phase II. J Clin Oncol 34(Suppl; abstr 1080)
55. Litton JK, Scoggins M, Ramirez DL, Murthy RK, Whitman GJ, Hess KR, Adrada BE, Moulder SL, Barcenas CH, Valero V, Booser D, Gomez JS, Mills GB, Piwnica-Worms H, Arun BK (2016) A pilot study of neoadjuvant talazoparib for early-stage breast cancer patients with a BRCA mutation. Ann Oncol 27(Suppl 6):vi43–vi67
56. Jaspers JE, Kersbergen A, Boon U, Sol W, van Deemter L, Zander SA, Drost R, Wientjens E, Ji J, Aly A, Doroshow JH, Cranston A, Martin NM, Lau A, O'Connor MJ, Ganesan S, Borst P, Jonkers J, Rottenberg S (2013) Loss of 53BP1 causes PARP inhibitor resistance in Brca1-mutated mouse mammary tumors. Cancer Discov 3(1):68–81. https://doi.org/10.1158/2159-8290.CD-12-0049
57. Xu G, Chapman JR, Brandsma I, Yuan J, Mistrik M, Bouwman P, Bartkova J, Gogola E, Warmerdam D, Barazas M, Jaspers JE, Watanabe K, Pieterse M, Kersbergen A, Sol W, Celie PH, Schouten PC, van den Broek B, Salman A, Nieuwland M, de Rink I, de Ronde J, Jalink K, Boulton SJ, Chen J, van Gent DC, Bartek J, Jonkers J, Borst P, Rottenberg S (2015) REV7 counteracts DNA double-strand break resection and affects PARP inhibition. Nature 521(7553):541–544. https://doi.org/10.1038/nature14328
58. Pettitt SJ, Rehman FL, Bajrami I, Brough R, Wallberg F, Kozarewa I, Fenwick K, Assiotis I, Chen L, Campbell J, Lord CJ, Ashworth A (2013) A genetic screen using the PiggyBac transposon in haploid cells identifies Parp1 as a mediator of olaparib toxicity. PLoS One 8(4):e61520. https://doi.org/10.1371/journal.pone.0061520
59. Chaudhuri AR, Callen E, Ding X, Gogola E, Duarte AA, Lee JE, Wong N, Lafarga V, Calvo JA, Panzarino NJ, John S, Day A, Crespo AV, Shen B, Starnes LM, de Ruiter JR, Daniel JA, Konstantinopoulos PA, Cortez D, Cantor SB, Fernandez-Capetillo O, Ge K, Jonkers J, Rottenberg S, Sharan SK, Nussenzweig A (2016) Erratum: replication fork stability confers chemoresistance in BRCA-deficient cells. Nature 539(7629):456. https://doi.org/10.1038/nature19826
60. Edwards SL, Brough R, Lord CJ, Natrajan R, Vatcheva R, Levine DA, Boyd J, Reis-Filho JS, Ashworth A

(2008) Resistance to therapy caused by intragenic deletion in BRCA2. Nature 451(7182):1111–1115. https://doi.org/10.1038/nature06548
61. Barber LJ, Sandhu S, Chen L, Campbell J, Kozarewa I, Fenwick K, Assiotis I, Rodrigues DN, Reis Filho JS, Moreno V, Mateo J, Molife LR, De Bono J, Kaye S, Lord CJ, Ashworth A (2013) Secondary mutations in BRCA2 associated with clinical resistance to a PARP inhibitor. J Pathol 229(3):422–429. https://doi.org/10.1002/path.4140
62. Sakai W, Swisher EM, Karlan BY, Agarwal MK, Higgins J, Friedman C, Villegas E, Jacquemont C, Farrugia DJ, Couch FJ, Urban N, Taniguchi T (2008) Secondary mutations as a mechanism of cisplatin resistance in BRCA2-mutated cancers. Nature 451(7182):1116–1120. https://doi.org/10.1038/nature06633
63. Lord CJ, Ashworth A (2016) BRCAness revisited. Nat Rev Cancer 16(2):110–120. https://doi.org/10.1038/nrc.2015.21
64. McCabe N, Turner NC, Lord CJ, Kluzek K, Bialkowska A, Swift S, Giavara S, O'Connor MJ, Tutt AN, Zdzienicka MZ, Smith GC, Ashworth A (2006) Deficiency in the repair of DNA damage by homologous recombination and sensitivity to poly(ADP-ribose) polymerase inhibition. Cancer Res 66(16):8109–8115. https://doi.org/10.1158/0008-5472.CAN-06-0140
65. Gilardini Montani MS, Prodosmo A, Stagni V, Merli D, Monteonofrio L, Gatti V, Gentileschi MP, Barila D, Soddu S (2013) ATM-depletion in breast cancer cells confers sensitivity to PARP inhibition. J Exp Clin Cancer Res 32:95. https://doi.org/10.1186/1756-9966-32-95
66. Subhash VV, Tan SH, Yeo MS, Yan FL, Peethala PC, Liem N, Krishnan V, Yong WP (2016) ATM expression predicts Veliparib and Irinotecan sensitivity in gastric cancer by mediating P53-independent regulation of cell cycle and apoptosis. Mol Cancer Ther 15(12):3087–3096. https://doi.org/10.1158/1535-7163.MCT-15-1002
67. Villalona-Calero MA, Duan W, Zhao W, Shilo K, Schaaf LJ, Thurmond J, Westman JA, Marshall J, Xiaobai L, Ji J, Rose J, Lustberg M, Bekaii-Saab T, Chen A, Timmers C (2016) Veliparib alone or in combination with Mitomycin C in patients with solid tumors with functional deficiency in homologous recombination repair. J Natl Cancer Inst 108(7). https://doi.org/10.1093/jnci/djv437