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**PARP inhibitors: an interesting pathway also for non-small cell lung cancer?**

*Giaj Levra M, Olaussen KA, Novello S, Soria JC*

***Abstract**: Treatment of lung cancer is improving, also based on the identification of molecular characteristics of the tumor, of which some already constitute promising targets. One of the molecular characteristics thought to play an important role in lung cancer is DNA repair dysfunctionality. Deregulated expression of DNA repair proteins, such as PARP, has been studied in lung cancer as a possible biomarker and clinically useful target, but the literature remains relatively poor. Pharmacological inactivation of PARP has allowed the, identification of a synthetic lethality with a second DNA repair protein such as BRCA1, but has also shown the potential to sensitize tumors to commonly used cytotoxic agents. The current manuscript reviews data regarding PARP in the context of DNA repair and its different pathways, as well as the clinical data generated until now with PARP inhibitors. A deeper understanding of the DNA damage response in lung malignancies, and particularly a clarification of the crosstalk between DNA repair functionality and genetic stability, is the key to optimize the development of PARP inhibitors in the setting of NSCLC.*

**Keywords**: PARP1, lung cancer, synthetic lethality

**INTRODUCTION**

Damaged DNA and subsequent genomic instability accompany the malignant transformation of cells and subsequent metastatic processes. In parallel, a number of tumors are characterized by the loss of functionality in one or several DNA repair pathways, such as in a subset of breast and ovarian cancers (BRCA1 or BRCA2 repression/mutation, which are essential genes in the homologous recombination repair pathway), in colon cancer (methylated promoter/loss of hMLH1 or MSH2 expression, essential proteins in the mismatch repair pathway), in gliomas (methylated promoter/loss of MGMT expression) and in lung cancer (loss of ERCC1 expression).

Damaged DNA is signaled through a DNA Damage Response (DDR) mechanism allowing cell cycle regulation, apoptotic regulation and/or a rapid processing of insults by the DNA repair machinery. Poly-ADP ribosylation (PAR) is a post-translational modification of proteins or histones that is catalyzed mainly by the poly- ADP ribose polymerase 1 (PARP1) and PARP2. It is believed that the PAR process is an important early cellular response to DNA damage. The process causes chromatin decondensation around the damage sites.

This “activation” allows the recruitment of repair machineries such as the base excision repair (BER) complexes. The system thereby accelerates DNA repair, particularly in the case of single-stranded breaks (SSBs) [1]. PARP1 also seems to affect double-strand break (DSB) repair, since PARP1 deficient cells are hypersensitive to DSB inducing agents. Indeed, PARP is activated at stalled replication forks allowing Homologous Recombination Repair (HRR) and mediates Mre11-dependent replication restart [2].

In this review, we enumerate the different PARP inhibitors that have been evaluated clinically in lung cancer. Compared to breast and ovarian cancers, the idea of using PARP inhibitors in lung cancer is not related to BRCA1/2 mutations, since these are not found. However, in tumors carrying intrinsically high genomic instability (or high DNA repair dysfunctionality), targeting PARP might induce a “saturation” of the systems controlling DNA damage with subsequent cell death. A second strategy has been to evaluate the combination of PARP inhibitors with DNA interacting chemotherapeutic agents such as platinum compounds. Indeed, pharmacological PARP inhibition increases platinum adduct accumulation in NSCLC cells suggesting a slow-down of platinum DNA-adduct repair capacity, which eventually potentiates the efficacy of platinum- based chemotherapy independently from p53-status [3,4].

## PARP IN THE CONTEXT OF SINGLE STRAND BREAK REPAIR

PARP is an essential protein in the BER pathway, which is responsible of the removal of small (non bulky) base damages with subsequent single strand breaks (SSBs). These damages on one base of the DNA might originate from endogenous insults (mainly oxidative modifications, but also deamination or hydrolysis of glycosyl DNA bonds leading to loss of the entire base) or have an exogenous origin (X-rays, monofunctional alkylating agents, or antimetabolites) [5]. The initial step of BER is the removal of the damaged base by a DNA glycosylase, which induces the formation of an abasic (apurinic/apyrimidinic) site [6]. The abasic site is recognized by APE1 endonuclease in association with PARP and XRCC1. PARP-mediated ribosylation largely modifies the structure of histones and other proteins surrounding the lesion [7]. The abasic site is processed by APE1, whereas XRCC1, which is one of the acceptor proteins of the ADP-ribosyl polymers, recruits DNA polymera **β** that fills in the lacking single nucleotide (short patch BER). Less frequently, a longer 2-15 nucleotides repair is involved (long-patch BER) with the contribution of other proteins such as PCNA and FEN1. In both long and short patch BER, the final ligation is performed by DNA ligase III. Noteworthy, recent work has linked the BER pathway to the nucleotide excision repair (NER) pathway with the implication of DNA structure-specific nucleases such as ERCC1 [8,9].

The role of PARP in BER-mediated SSB repair appears as essential. PARP is rapidly recruited to damaged DNA thereby activating its catalytic domain that catalyze both the covalent transfer of the ADP-ribose moiety of nicotinamide adenine nucleotides (NAD+) to a variety of nuclear protein acceptors, a reaction that consumes NAD+ and thereby ATP (Fig. 1) [10]. The reaction creates branched polymers of 60 to 80 ADP-ribose units on PARP itself that PARP transfers to other acceptor proteins such as XRCC1. Since the polymers of ADP-ribose units are negatively charged, they cause an electrostatic repulsion between poly-ADP ribosylated proteins and DNA with a subsequent local relaxation of DNA structures [11]. This phenomenon is thought to improve the accessibility of DNA to repair enzymes [12].

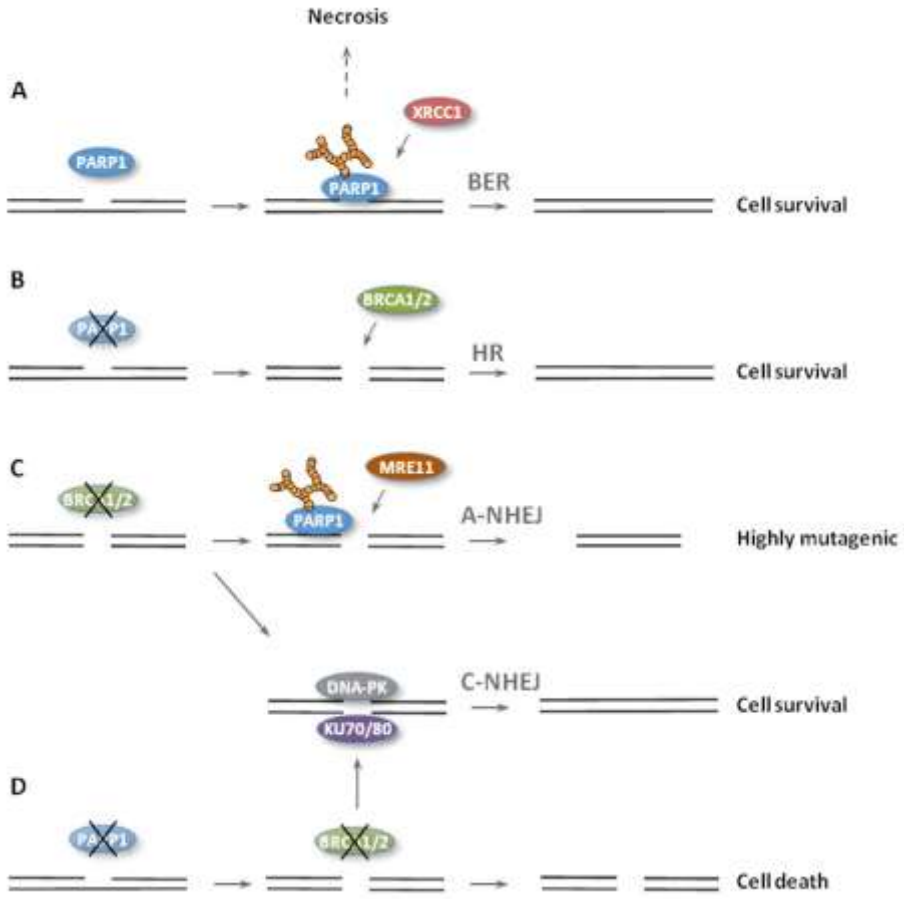


*Fig. (1). Function of PARP and consequences of PARP inhibitors. Upon spontaneous or induced DNA single strand breaks or other therapeutically induced lesions (interstrand cross-links are not shown for the sake of clarity), PARP1 is recruited to DNA where it immediately polymerizes branched ADP-ribosyl units on itself, which allows the recruitment of DNA repair proteins such as XRCC1 in the base excision repair (BER) pathway (A). If the level of DNA damage is very high, the subsequent NAD/ATP depletion due to Poly(ADP-ribosyl)ation will trigger necrotic cell death. The use of PARP inhibitors in cancer cells eventually leads to DSBs during replication. These DSBs are repaired by the homologous recombination (HR) pathway in cells containing active BRCA1/2 proteins (B) or by non-homologous end joining (NHEJ). The balance between HR and NHEJ pathway largely depends on AKT phosphorylation. Cells with inactive or mutant BRCA1/2 (also called "BRCAness") are unable to activate HR upon DSB damage (C). Instead, the DSBs are processed by either the nonmutagenic canonical NHEJ (C-NHEJ) pathway or the highly mutagenic alternative NHEJ (A-NHEJ) pathway (also known as microhomology-mediated end joining MMEJ). The subsequent genetic instability is probably one of the drivers of carcinogenesis in cells with homozygous BRCA1/2 mutations. Nuclease activity of MRE11 favors A-NHEJ and requires the presence of PARP. The use of PARP inhibitors in cells with no or mutant BRCA1/2 leads to synthetic lethality of cancer cells (D). At least theoretically, this should particularly be true in cells with repressed C-*

*NHEJ pathway due to inactivation of proteins such as DNA-PK, KU70/80, Artemis, XRCC4, Ligase IV or to blocked AKT signaling.*

**THE FAMILY OF PARP PROTEINS AND ALTERNATIVE ROLES**

Nuclear DNA-interacting proteins that are targets of poly-ADP ribosylation include core histones H1 (chromatin relaxation), HMG proteins, topoisomerases I and II (genomic maintenance), WRN (several DNA repair pathways), DNA-PKcs/Ku70/Ku80 complex (NHEJ DNA repair pathway), ATM (HR pathway and checkpoint activation), MRE11 (HR pathway and restarting of the collapsed replication fork), and the well-known XRCC1/DNA ligase IIIlalpha/PolBeta complex (BER pathway).

Since several proteins are targeted by poly-ADP ribosylation, PARP also participates in other key cellular functions than simply being DNA damage "sensors". These other functions are cell death programs (PARP is cleaved by caspase-3 through the AIFdependant apoptosis pathway), transcription regulation, telomere cohesion and mitotic spindle formation during cell division (fall in ATP prevents cells with unrepaired DNA damaged from entering mitosis), intracellular energy metabolism, and intracellular trafficking of proteins such as p53, NF-κB and HMGB1 [13]. The many alternative functions are partly explained by the existence of least 17 members located on different regions of the human genome. Further complexity of PARP related functions has been explained by alternatively spliced transcript variants encoding different mRNA isoforms of several of the gene members. At least six of the PARP members are structurally related and subdivided in 3 groups, with PARP1, 2 and 3 in the first group,

PARP4 in the second and tankyrase 1 (TKNS) and TKNS2 in the third [14]. PARP1 and PARP2 are the most abundant proteins and represent the majority of large PAR activity in human cells. PARP1 and PARP2 are therefore the two most studied of all the PARP genes.

PARP1 is considered to best characterize the activation of the BER pathway. The PARP1 gene is located on chromosome 1q43 and codes a protein whose molecular weight is 116 kDa. The PARP1 protein carries 3 main domains, one N-terminal zinc-finger domain that detects and binds to DNA single strand breaks, one selfmodification inhibitory domain that carries poly(ADP)- ribosylation sites, and one C-terminal catalytic domain [9,15,16].

PARP2 corresponds to the C-terminal part of PARP1 (catalytic domain binding the NAD+), but it does not possess a Zn finger DNA binding domain and the central selfmodification domain found in PARP1. Although accounting for only a small part of the total poly-ADP ribosylation stimulated by DNA SSBs, PARP2 plays a role in genomic integrity maintenance as it interacts with PARP1 as a hétérodimère. Therefore, both proteins can modify themselves mutually through poly-ADP ribosylation. Interestingly, cells from PARP2-deficient mice treated with alkylating agents displayed delayed DNA strand break resealing similar to that observed in PARP1-deficient cells [17].

**CONSEQUENCE OF PARP INHIBITION**

Since PARP proteins act as DNA damage "sensors" that allows modulation of BER capacity, PARP is a potential target for inhibition of DNA repair (Fig. 1). Modulation of poly-ADP ribosylation is therefore considered a promising approach in clinical practice [18]. As stated earlier, the loss or repression of PARP1 expression is sufficient to induce cell death in cancer cells bearing BRCA1 or BRCA2 homozygous mutations, which are rate-limiting genes in the HRR pathway [19]. This observed synthetic lethality has lead to innovative clinical perspectives. PARP inhibition also suggests a
promising approach to sensitize tumor cells to chemo- and radiotherapy.
The inactivation of PARP in mice has lead to increased sensitivity to ionizing radiation and alkylating agents and to genomic instability [15]. Although the mechanism is not fully understood, the affinity of PARP1 for platinum-modified DNA *in vitro* has been established using modified cisplatin analogues to synthesize 25 bp DNA duplexes carrying platinum intrastrand crosslinks [20]. The activation of PARP upon cisplatin treatment is poorly characterized, but PARP could facilitate the dissociation of nuclear proteins from platinum-modified DNA that in turn could facilitate
recruitment of DNA repair proteins [21]. Recent work has shown that PARP1 inhibition influences DNA repair capacity of DNA platinum adducts with subsequent synergy with cisplatin treatment by inducing the intrinsic pathway of apoptosis [3,22]. The hypothesis that PARP1 is implicated in cisplatin resistance was further strengthened by recent work showing that PARP1 is highly expressed and constitutively hyperactivated in human ciplastinresistant cancer cells [4]. These cells with high levels of poly(ADPribosyl) ated proteins (high PARP1 activation) strongly induced apoptosis in response to pharmacologic PARP inhibitors suggesting that the response to PARP inhibitors can, in several cases of platinum-refractivity, be predicted by the level of PAR activity. PARP inhibitors also have the capacity to radiosensitize cancer cells [23]. Further, the global expression pattern of the DNA repair proteins, such as PARP might have predictive implications in the adjuvant setting of NSCLC treatments. Interestingly, one study has linked a single nucleotide polymorphism (SNP) in the PARP1 gene with response of non-small cell lung cancers (NSCLC) to chemotherapy [24]. Our own recent studies have suggested that PARP1 tumor positivity might constitute a molecular context with theranostic interest when it is combined with ERCC1 and MSH2 status in NSCLC, with histological subtypes as possible subgroups that should be studied separately [3,25]. Altogether, targeting PARP could become of large interest in treatment schedules of NSCLC. A valid definition of a "BRCAness" phenotype in NSCLC would help identifying patients who may respond optimally to PARP inhibitors.

**PARP INHIBITORS IN THE CLINIC**

Despite a large number of chemically distinct PARP inhibitors synthesized to date and explored for their putative therapeutic utility,only six have been tested in NSCLC (Table 1). Initially, preclinical studies suggested the potential efficacy of therapy combining PARP inhibitors with cisplatin in hepatocellular cancer [26]. In triple-negative breast cancer PARP inhibitors synergized with carboplatin plus gemcitabine [27,28]. PARP inhibitors that have been tested clinically in solid tumors as either monotherapy or in combination are olaparib (AZD2281), veliparib (ABT-888), iniparib (BSI-201), AG-014699, PF01367338, MK-4827, and CEP-9722, and EISAI.

**Olaparib**

Olaparib (AZD2281, Astra Zeneca/ KuDOS) is an orally active PARP inhibitor. Initial *in vitro* study showed its activity in association with the alkylating agent methanesulphonate (MMS) and *in vivo* study confirmed the ability of the molecule to potentiate the antitumor activity of methylating chemotherapeutic agents as temozolomide without exacerbation of the systemic toxicity. BRCA deficient cell lines were hypersensitive to olaparib in comparison with BRCA 1 and 2 proficient cells [29]. The efficacy of the molecule in BRCA mutated tumors was reported in a phase I study where 60 patients with solid tumors were enrolled; initially the study inclusion criteria didn't require the presence of the mutation, while in the expansion phase only patients with a BRCA mutation were enrolled (n=22). The maximum administered dose of olaparib was 600 mg twice daily and the maximum tolerated dose was 400 mg twice daily. The pharmacokinetic study showed a rapid absorption of the drug (peak of plasma concentration observed between 1 and 3 hours) and a rapid elimination with a half life of approximately 5 to 7 hours. The most common adverse events were nausea, fatigue and vomiting, with a limited incidence of myelosuppression. No obvious increase in the frequency of grade of adverse events was observed comparing known BRCA1 or BRCA2 mutation carriers with non carriers. A durable antitumor activity was observed in patients with a BRCA1 or BRCA2 mutation, or in patient with a
family history of BRCA mutation: of the 19 BRCA carriers evaluable patients, 12 (63%) had a clinical benefit with a radiologic or tumor-marker response, while no objective antitumor responses were observed in patient without known BRCA mutations [30]. A multicenter, single arm, phase II study sought to test the efficacy of olaparib in refractory breast cancer with a confirmed BRCA1-2 mutation and to evaluate the safety and the tolerability of the molecule. Such study was conducted in 54 patients (27 treated with olaparib 400 mg bid, 27 at the dose of 100 mg bid) with an
objective response rate (ORR) of 41% and 22% respectively. PFS was 5.7 months in the cohort with 400 mg bid and 3.8 in the cohort with 100mg bid [31,32]. A good tolerability of the treatment was reported with grade 3 or higher toxicities seen in 5 patients in terms of fatigue, nausea and anemia.

The same study design was applied for a phase II study of olaparib in patients with refractory BRCA mutated ovarian cancer [33]. The first cohort (33 patients) was given daily olaparib at the dose of 400 mg bid, and the second cohort (24 patients) was given continuous olaparib 100 mg bd. The ORR was 33% in the arm with 400 mg bid and 13% in the 100mg arm; the most frequently reported adverse events were nausea and fatigue. CTCAE grade 3 or 4 events were reported in 17 of 33 patients in the olaparib 400 mg cohort and in 14 of 24 patients in the olaparib 100 mg cohort.
The ICEBERG-3 study is a phase II open-label study, comparing the efficacy of olaparib to pegylated liposomial doxorubicin in ovarian cancer carrying the BRCA mutation that has recently completed the enrollment [34].

**Table 1. PARP inhibitors in the clinic**

| | Olaparib AZD2281 | Veliparib ABT-888 | Iniparib BSI-201 | AG-014699 PF01367338 | MK-4827 | CEP-9722 |
|---|---|---|---|---|---|---|
| **Pharma** | KuDos / AstraZeneca | Abbott | BiPar / Sanofi-Aventis | Pfizer/ Clovis Oncology | Merck | Cephalon |
| **Mechanism** | NAD competitive | NAD competitive | Zn Finger? | NAD competitive | NAD competitive | NAD competitive |
| **PARP1 IC50 (nM)** | 5 | 5 | Not reported (Not a real PARP inhibitor) | 1,4 Ki | 4 | 20 |
| **PARP2 IC50 (nM)** | 1 | 2 | Not reported (Not a real PARP inhibitor) | 0,2 Ki | 2 | 6 |
| **Route** | Oral | Oral | IV | IV | Oral | IV |
| **Use** | Mono / Combo | Mono / Combo | Combo | Mono / Combo | Mono | Combo |
| **Status** | Ph. 3 | Ph. 2 | Ph. 3 | Ph. 2 | Ph. 1 | Ph. 1 |

Olaparib is also tested in the maintenance setting; a phase II randomized trial of olaparib monotherapy as maintenance therapy after second-line platinum based therapy in platinum-sensitive ovarian cancer has recently completed accrual and preliminary data were presented at the ASCO meeting 2011: the PFS by RECIST criteria was significantly longer in the olaparib 400 mg bid than the placebo group (HR, 0.35; 95% CI 0.25–0.49; P<0.00001; median 8.4 vs 4.8 months), with a toxicity profile similar to the previous studies [35].

**Iniparib**

Iniparib (BSI-201) is a small molecule with very weak PARPinhibitory properties, and which real mechanism of action is yet to be elucidated [36]. The safety, tolerability and efficacy of BSI-201 in monotherapy or in association with other chemotherapic agents in solid tumors (topotecan 1.5 mg/mq; gemcitabine 1000 mg/mq; temozolomide 75 mg/mq or carboplatin AUC6 and paclitaxel 200 mg/mq) was evaluated in different phase I trials [37,38]. In all the studies the treatment with BSI-201 was well tolerated with a maximum dose of BSI-201 administered of 8.0 mg/kg twice

weekly; in the Mahany's study all the patients tolerated doses up to 5.6 mg/kg with two serious adverse events possibly related to iniparib. A randomized phase II study in triple negative breast cancer evaluated the efficacy of carboplatin AUC2 and gemcitabine 1000 mg/mg (administered on days 1 and 8 every 21 days) alone or in association with BSI-201 (at 5,6 mg/kg e.v. biweekly on days 1, 4, 8 and 11); the trial showed an efficacy of the experimental arm, with a clinical benefits rate of 12% for the control arm and 52% for the experimental arm (clinical benefits encompassed a complete response, a partial response, or a stable disease). The progression free survival was 6,9 months for the experimental arm and 3,3 months for the control arm (p<0.0001, HR=0,342 CI 0.2-0.584) and overall survival 9,2 versus 5,7 months (p=0.0005, HR= 0,348 CI 0,189-0,649) [39]. In December 2009 at the San Antonio Breast Cancer Symposium results updated showed an overall survival of the chemotherapy arm of 7,7 months and 12,2 for the association arm (p= 0.005; HR= 0,5; 95% CI 0.30-0.82) [40]. According to these data a phase III trial to evaluate the efficacy of iniparib in association with chemotherapy in patients with triple negative breast cancer previously treated or in first line was conducted. The results were presented in 2011 at the ASCO meeting in Chicago and the key end points were not met: the PFS was 5,1 months for the patients in the experimental arm and 4,1 months in the control arm (p= 0,027; HR: 0,79; 95% CI 0,646-0,976) and the OS was 11,8 versus 11,1 months (p= 0,28; HR: 0,87; 95% CI 0,687- 1,116). No differences were observed in terms of response rate between the two arms and the study confirmed the good profile of tolerability and toxicity of iniparib. An exploratory analysis showed a possible benefit for the use of iniparib in association with chemotherapy in second and third line [41].

Iniparib appeared to potentiate the anti-tumor activity of chemotherapy also in patients with ovarian cancer, either sensitive or not to cisplatin and two abstracts were presented at the ASCO meeting in 2011. A phase II study in platinum sensitive patients, demonstrated that the combination of carboplatin (AUC 4 d1), gemcitabine (1000 mg/mq d1,8) and iniparib (5,6 mg/Kg d1,4,8,11) achieved an overall response rate of 70,6%, that is better than the 47% reported in literature [42,43]. In the second phase II trial in patients with platinum resistant ovarian cancer the association of iniparib with the chemotherapy showed a progression free survival and an overall response rate in line with those achieved with the pegylated liposomal doxorubicine (ORR: 31,6% versus 11,7%; PFS: 5,9 versus 3,1 months) [44,45].

**Other PARP Inhibitors**

The first phase I in human of a PARP inhibitor was the molecule AG014699 (Pfizer) in association with temozolomide in a total of 33 patients with solid tumors. In the first part of the trial the primary objective was to find the PARP inhibitory dose (PID), based on 74-97% inhibition of peripheral blood lymphocyte (PBL). The PID was 12 mg/mq and no dose-limiting toxicities were observed [46,47]. Subsequently, AG014699 was tested with temozolomide 200 mg/mq daily for 5 days every 4 weeks; a further dose increase of AG014699 to 18 mg/mq was evaluated without any

additional evidence of PARP inhibition, however the dose was intolerable with a significant rate of myelosuppresion. Responses were seen in patients with melanoma, desmoids tumors, pancreas cancer, leyomiosarcoma and prostate cancer.

The phase II study of AG014699 12 mg/mq with temozolomide 200 mg/mq in patient with metastatic melanoma showed a partial response rate of 18% and a prolonged disease stabilization (>6 months); however an increased frequency of myelosuppresion was found, with a 12-15% of cycles with grade 4 neutropenia and thrombocytopenia reported, necessitating a 25% dose reduction [48]. A phase II of AG014699 in combination with cisplatin in triple negative breast cancer is ongoing.

Veliparib (ABT-888; Abbott Laboratories) is the first compound tested in a phase 0 clinical trial with target modulation as primary end point in patients with advanced tumors; trial objectives were to evaluate its pharmacokinetic and the pharmacodynamic and to determine a dose range at which ABT-888 inhibits PARP in tumor tissue and PBMC. The study demonstrated that ABT-888 is rapidly absorbed, with a peak plasma level between 0.5 and 1.5 hours after dosing, with a clearance after 24 h of dosing. PARP inhibition of >85% lasting up to 24h post drug administration was observed in tumor tissue and PBMC at the dose of 25 and 50mg [49].

Several phase I studies investigated the DLT and the MTD of veliparib monotherapy or in combination with chemotherapic regimes (carboplatin and paclitaxel, carboplatin and gemcitabine or carboplatin monotherapy) in patients with advanced solid tumors (included NSCLC) or with BRCA-associated tumors, with the evidence of a well-tolerated drug with a safety profile [50-54].

MK-4827 (Merck) is a selective PARP 1-2 inhibitor tested in a phase I clinical trials in patients with BRCA-deficient and sporadic ovarian cancers. Dose-limiting toxicity was observed in 3 patients (grade 3 fatigue, pneumonitis and anorexia) and the antitumor activity was observed both in sporadic and BRCA-deficient cancers [55].

Two clinical trials are still ongoing with MK-4827, one is a phase I study in patients with solid tumors, and the other in association with temozolomide in advanced cancers. Eisai PARP inhibitor E7016 was reported at the EORTC-NCI-AACR 2012 meeting in combination with Temozolomide [56].

**PARP INHIBITORS AND NSCLC**

A phase Ib study, presented at the ASCO meeting 2011, evaluating the activity of iniparib 5,6 mg/kg d1,4,8,11 in association with carboplatin AUC 6 d1 and paclitaxel 200 mg/mq d1 in first line treatment, showed a promising activity of this combination, with an increased overall response rate compared with previous studies with carboplatin and paclitaxel alone (20% versus 15%) in patients with stage IIIB-IV non-small cell lung cancer (NSCLC) . Three patients had a confirmed partial response, 9 patients showed a stable disease (6 through 12 weeks and 3 through 18 weeks) with only a progressive disease. The most common toxicities were grade 1-2 asthenia (40%),

anemia (33,3%) and nausea (26,7); grade 3-4 adverse events were exclusively neutropenia (13,3%) and anemia (6,7%) [57].

At the IASLC (International Association for the study of Lung Cancer) meeting in Amsterdam in 2011 the preliminary results of a randomized (2:1), open label phase II trial of gemcitabine 1250 mg/mq (d1 and 8) in association with cisplatin 75 mg/mq (d1) with or without iniparib 5,6 mg/kg (d 1,4,8,11) in first line treatment in patients with NSCLC were presented (Fig. 2). One hundred and nineteen patients were enrolled, 80 treated in the experimental arm and 39 in the control one; primary end-point was the ORR, secondary: PFS, OS, and safety of this combination. The overall response rate for the iniparib arm was 21% (with one case of complete response) and 26% for the control arm and no differences in response by subgroups were seen. The preliminary results of progression free survival showed a not statistically significant trend in favor of the experimental arm with iniparib (5,7 vs 4,3 months, p: 0,145, HR: 0,69, 95% CI: 0,43-1,14); while the toxicity profile of the combination of cisplatin, gemcitabine and iniparib was similar to the one observed with cisplatin and gemcitabine alone, with asthenia, nausea and neutropenia as most common averse events (neutropenia was the most frequent grade 3-4 adverse events in both arms). The authors recommended caution in the interpretation of the advantage in terms of progression free survival, because the two groups were imbalanced in performance status and gender distribution [58].

Mature results for overall and progression free survival will be presented soon. The ECLIPSE study was a phase III trial to evaluate (as primary end point) the overall survival (OS) of patients with advanced squamous cell lung cancer receiving a combination of carboplatin gemcitabine either with or without BSI-201 in first line (Fig. 3). Eight hundred twenty-five patients were planned to be treated with a 1:1 randomization schema and they were planned to receive carboplatin AUC5 d1-gemcitabine 1000 mg/mq d 1,8 either with or without iniparib 5,6 mg/kg d1,4,8,11 every three weeks. Secondary endpoints of the study were: progression free survival (PFS), time to progression (TTP), overall response rate (ORR) and safety and tolerability of the treatment. A recent press release announced that the trial did not achieve an improvement in overall survival and there were no meaningful differences in the main safety parameters between the two arms [59]. Two clinical trials are evaluating the role of olaparib in patients with NSCLC: one is a phase I trial of olaparib in association or not with cisplatin and concurrent radiotherapy in locally advanced disease, the other is a randomized phase Ib/II open label study, designed to evaluate the efficacy and tolerability of gefitinib with or without olaparib in patients with NSCLC harboring the Epidermal Growth Factor Recptor (EGFR) mutation (Table 2). Veliparib (ABT-888) is under investigation in NSCLC in three ongoing clinical trials: one is phase I-II trial evaluating the DLT and PFS of veliparib in association with carboplatin/paclitaxel, with or without radiation therapy in patients with stage III NSCLC that cannot underwent surgery, the second one is a phase I trial with cisplatin and gemcitabine in NSCLC, biliary, pancreatic or urothelial cancer.

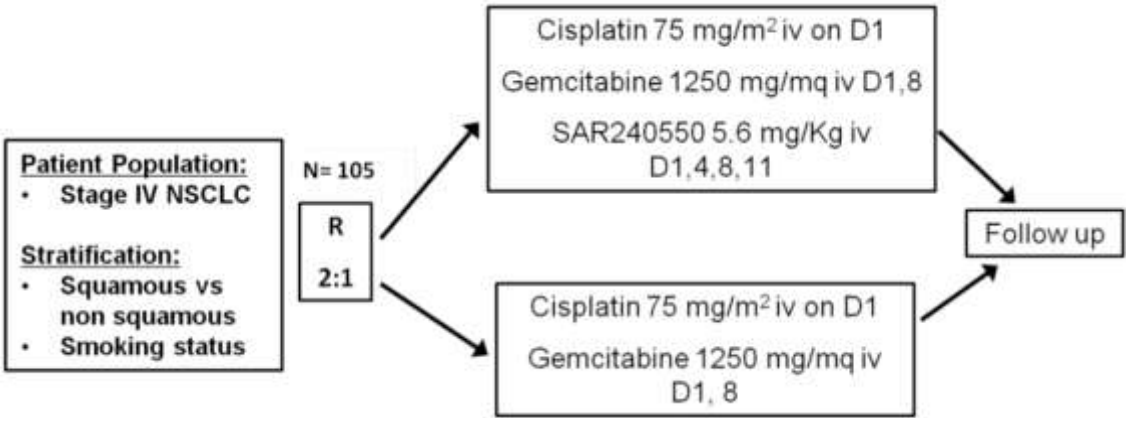


Fig. (2). Study design of the phase II trial of iniparib in NSCLC.

**Table 2. Clinical trials published or ongoing with PARP inhibitors in Non Small Cell Lung Cancer**

| Product | Clinical trial | Development |
|---|---|---|
| Iniparib (BSI-201) | NSCLC in association with carboplatin and paclitaxel | Ib |
| | NSCLC in association with cisplatin and gemcitabine | II |
| | Squamous cell lung carcinoma, in association with carboplatin and gemcitabine | III |
| Olaparib (AZD2281) | In patients with the Epidermal Growth Factor Receptor (EGFR) mutation in combination with gefitinib versus gefitinib alone | I-II |
| | Locally advanced NSCLC in association with radiation therapy with or without cisplatin | I |
| ABT-888 | Stage III NSCLC in combination with carboplatin, paclitaxel with or without radiation therapy | I-II |
| | NSCLC in association with cisplatin and gemcitabine | I |
| | NSCLC with brain metastases in combination with radiotherapy versus placebo plus radiotherapy | II |
| | NSCLC, in association with carboplatin and paclitaxel | II |

A randomized phase II, double-blind, dose ranging trial is evaluating the safety and the efficacy of veliparib and whole brain radiation therapy in subjects with brain metastases from NSCLC (Table 2).

## CONCLUSION

Results from preclinical studies and first clinical trial showed promising data about the possible role of PARP inhibitors in association with standard chemotherapies regimes. However, recent results have challenged the PARP-inhibitory nature of iniparib and the PK profile of olaparib is currently been reassessed. Preliminary data in lung cancer showed an activity of iniparib in association with cisplatin/gemcitabine, but not a statistically advantage in terms of progression free survival; more information will be available deriving from the final results of the phase III trial, although preliminary data are not completely encouraging.

Many questions are still open, such as the potential patients that could more benefit from these molecules, the line of therapy in which these drugs can be used and the optimal chemotherapy regimens associated. A major challenge in the next years is also to identify the presence of tumors with a BRCAness features, that potentially could benefit of this treatment together with the discovery of new predictive biomarkers.

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