HHS Public Access

Author manuscript

Biochem Soc Trans. Author manuscript; available in PMC 2020 July 02.

Published in final edited form as:

Biochem Soc Trans. 2020 June 30; 48(3): 1101–1108. doi:10.1042/BST20191092.

Systems approach to rational combination therapy: PARP inhibitors

Chaoyang Sun[1], **Yong Fang**[2], **Marilyne Labrie**[2], **Xi Li**[2], **Gordon B. Mills**[2]

[1]Department of Obstetrics and Gynecology, Tongji Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan 430030, China.

[2]Department of Cell, Development and Cancer Biology, Knight Cancer Institute, Oregon Health and Sciences University, Portland, OR 97201, USA,

Abstract

Poly (ADP-ribose) polymerase inhibitors (PARPi) have demonstrated activity across a broad spectrum of molecular backgrounds and tumor types, with the greatest activity observed in patients with aberrations in the homologous recombination DNA damage repair pathway. Despite remarkable responses in a subset of patients, the response is usually modest and transient due to the almost inevitable emergence of resistance. Tumors develop resistance through rapid adaptation to the effects of PARPi as well as by generation or selection of genomic aberration. Although adaptive responses results in drug resistance, it also induces therapeutic vulnerabilities that could be exploited with rational combination therapies. To fulfill this role, we established the Combinatorial Adaptive Response Therapy (CART) platform by performing reverse-phase protein arrays (RPPA) to characterize adaptive responses, and develop rational combination therapies. Our series of studies strongly support the efficacy of this strategy, wherein targeting the emerging adaptive responses to PARPi with MEK/ERK inhibitors, WEE1/ATR inhibition (inhibitors of S phase and G2 DNA damage checkpoint), and PI3K/AKT/mTOR inhibition, and showed promising anti-tumor activity in various preclinical models. Importantly, this approach has been proven highly efficient, and several combinational therapies developed from the CART platform are being evaluated in ongoing clinical trials (NCT03801369, NCT03586661, NCT03162627, NCT03544125, NCT02659241, NCT02208375, NCT02316834, and NCT03637491).

Introduction

The poly (ADP-ribose) polymerase (PARP) family comprises a group of 17 members that play crucial roles in various aspects of DNA damage response (DDR)[1–3]. DDR involves

Author Contribution

C.S. and G.B.M. conceived and co-wrote the manuscript. Y.F. and M.L. provided precious comments and edited the manuscript.

Competing Interests

Gordon B. Mills

Scientific advisory board/Consultant: AstraZeneca, Chrysallis Biotechnology, ImmunoMET, Ionis, Lilly, PDX Pharmaceuticals, Signalchem Lifesciences, Symphogen, Tarveda,

Stock/Options/Financial: Catena Pharmaceuticals, ImmunoMet, SignalChem, Tarveda

Licensed Technology HRD assay to Myriad Genetics, DSP patents with Nanostring

Sponsored research: Nanostring Center of Excellence, Ionis (Provision of tool compounds only)

numerous intertwined signaling pathways responsible for preventing, detecting, and repairing DNA damage. The primary function of PARPs is to detect single-strand break (SSBs), recruit DNA repair complexes, and stabilize replication forks. Homologous recombination repair (HR) is the leading high fidelity DNA repair pathway for double strand breaks (DSB). The rationale of the anti-tumor effects of PARP inhibitors (PARPi) in HR defective (HRD) cancers induced by BRCA1 or BRCA2 mutations, is based on the idea that compromise in SSBs repair caused by PARPi would result in the accumulated redundancy of SSBs convert to DSBs, which induce cancer cells with HRD the synthetic lethality[1,4,5]. In contrast, normal cells retain the capacity to repair DSBs through HR and are therefore resistant to PARPi[4,5]. Following implementation into the clinic, it was found that single-agent PARPi abrogate PARP autoribosylation resulting in a “PARP trapping” phenomenon that can lead to catastrophic events in cancer cells via the accumulation of a DNA-PARP complex[1,6,7]. Further, PARP is required for the protection of the replication fork with PARP inhibitors causing replication fork collapse that leads to addition DNA damage and action of replication stress and the S phase checkpoint. Together, the SSB and DSB induced by PARPi activate the G2 checkpoint providing an opportunity for DNA repair. Both replication stress and the DNA damage mediated cell cycle blockade provide additional therapeutic opportunities.

Mounting clinical evidence has demonstrated the activity of PARPi across multiple diseases and treatment settings. Therefore, four PARPi received FDA approval for the treatment of different cancer types: olaparib, rucaparib, niraparib, and talazoparib in the ovary, breast, pancreas, and prostate cancers[1,8]. However, despite the success of PARPi monotherapy in cancer treatment, only a small subset of patients, even those with BRCA1 or BRCA2 mutations, have benefit from treatment of PARPi. Moreover, despite impressive responses were demonstrated in some patients, the response do not last long due to the almost inevitable development of drug resistance. As the emergence of resistance is the major challenge that limits duration and the magnitude of response to therapy, there is an urgent need to understand and halt PARPi drug resistance to improve patient outcomes. Adaptive responses, where the cancer cells and tumor ecosystem evolve fast by activating compensatory processes under therapeutic stress[9,10], allows tumors to survive the initial therapeutic stress caused by PARPi, leading to acquired resistance. Importantly, adaptive responses also give rise to therapeutic vulnerabilities, which can be used by simultaneously blocking adaptive rewiring pathways with rational combination therapies. Fortunately, although tumors evolve rapidly, there appears to be a limited “rewiring” ways for cancer cells to bypass survival stress induced by specific inhibitors including PARPi. Thus, if the rewired adaptive response pathways are discoverable, in turn, they could be taken advantage effectively to design rational combination therapies for particular patients.

DNA damage is a common cellular event induced by exogenous and endogenous stressors. Thus, cells have developed complex DDR mechanisms to limit and repair damage, maintaining the integrity of the genome. The interdiction of DNA repair by PARPi or toxins in the environment activates many pathways to avoid cell death to allow time for the cells to repair DNA damage. Indeed, the concept that cells must have developed approaches to deal with toxic stresses underlies systems biology theory and, in particular the concept of adaptive resistance. However, while almost every molecule in the cell must have been

targeted by mutation or stresses in the environment, such as by toxins in food or by infections, the chance that bypass survival pathways would be targeted by the same toxin is extremely unlikely. This underlies the overarching concept of combinatorial adaptive response therapy or CART as a therapeutic approach. Since the processes mediating DNA damage repair and maintenance of genomic integrity are robust when confronted by a single perturbation or therapeutic agent, rational combination therapy that targets the adaptive responses engendered by drugs, could delay or even overcome adaptive resistance and improve patient outcomes.

Thus we have developed the CART platform designed to exploit the adaptive response by which tumor ecosystems rapidly evolved to therapeutic stress induced by PARPis. For this purpose, we used a systematic approach to evaluate the adaptive responses, uncover vulnerabilities caused by therapy, and design rational combination therapies that can prevent or even reverse PARPi resistance and to expand the potential patient populations that might benefit from PARP inhibitors.

The CART platform

Proteins are the functional unit in the cell. We and others have demonstrated that protein levels and function are poorly corrected with DNA and RNA expressions[11–14]. Moreover, proteomic data includes essential information content, like protein post-translational modifications and functions of protein complexes, which are not available by analysis of DNA and RNA. Importantly, adaptive network rewiring mainly occurs at the protein level, supporting the need to identify protein alterations following drug treatment[9,10]. Thus, we have focused on proteomics-based technologies to study adaptive responses of tumors to PARPis. The RPPA technology is a high-throughput, semi-quantitative, and cost-effective approach for functional proteomics studies[11,12]. Up to now, we have validated almost 500 antibodies for RPPA, with 20% of proteins involved in post-translationally modifications, including phosphorylation, methylation, and histone acetylation. The proteins in critical oncogenic pathways were prioritised selected include DNA damage repair, replication stress, cell cycle progression, autophagy, proliferation, metabolism, apoptosis, cell surface tyrosine kinase receptors (TKI), PI3K/AKT/mTOR, RAS/RAF/MAPK, TGFα/β, and Wnt/beta-catenin and Hippo, YAP pathways. Accordingly, our Combinatorial Adaptive Response Therapy (CART) platform was implemented to characterize adaptive responses by using RPPA and subsequently design rational combination therapy by blocking critical signaling nodes or “rewired” signaling pathways. The inhibition of the rewired signaling pathways were expected to result in synthetic lethality with the original drug. Using this platform, we have explored adaptive responses in tumor and tumor ecosystems after treatment with multiple target therapies, including PARP, PI3K/AKT/mTOR, FAK, MEK/ERK inhibitors, EGFR antibodies, HDAC, BRD4, CHK1, WEE1, and ATR inhibitors. This platform is also applicable to other therapeutic options available to patients, including chemo and radiation therapies and immune checkpoint blockade, to identify rational combinations that could increase activity as well as overcome resistance. Furthermore, the platform is designed to identify biomarkers of sensitivity to monotherapies as well as drug combinations. Experimentally, highly characterized cancer cell lines (2D monolayer or 3D matrix-attached spheroid culture conditions), patient-derived xenograft (PDX) tumors from genetically

engineered mouse models (GEMM), or clinical patient tissues following various time-course and dose response to targeted therapies are comprehensively analyzed by RPPA. Then we quantify and visualize the collective adaptive resistance responses through individual proteins and signaling pathways, which provides a guideline to design and nominate rational combination therapies. Strikingly, targeting these acquired vulnerabilities showed an unprecedented efficacy rate as we have demonstrated in a number of studies[15–18]. In this paper, we will present the concepts of CART platform and illustrate proof of concept studies that support targeting adaptive responses induced by PARPi to increase the duration and depth of drug response.

Adaptive response after PARPi evaluated by RPPA

As we shown in Figure 1A, we applied the CART platform to dissect adaptive responses following PARP inhibition in ten well-characterized ovarian, endometrial, and breast cancer cell lines [IGROV-1 (BRCA1_Mut), SKOV3 (HER2_Amp, and PIK3CA_Mut), TOV21G (KRAS_Mut, and PIK3CA_Mut), ETN-1 (PTEN_Mut), KLE (TP53_Mut), HCC1954 (HER2_Amp, and PIK3CA_Mut), BT474 (HER2_Amp, and PIK3CA_Mut), SKBr3 (HER2_Amp), MDA-MB-468 (PTEN_Mut), and HCC1937 (BRCA1_Mut)] (Figure 1A). These cells were treated with two kinds of PARPi (AZD2281 and BMN673) under various conditions [two culture conditions (culture in 2D monolayer, or 3D matrix-attached spheroid), two dosages (IC25, and IC75), and two time-points (3 days, and 7 days) (Figure 1). Then cell lysates were evaluated by RPPA with 218 antibodies targeting total and phosphorylated proteins. In order to validate the generalization of the adaptive response and eliminate off-target effects after PARPi, we integrated results of protein alterations following both BMN673 and AZD2281 as we published previously[16,19]. First, we averaged the duplications under each treatment condition. Second, all protein expression levels following BMN673 and AZD2281 treatment at various conditions were normalized to its corresponding control (vehicle-treated cells at the same culture condition, time point, and dosages). Third, we added up each protein amounts for each cell line. Fourth, we rank-ordered protein alterations by median-centered scores to visualize the decreases or increases in the expression of proteins and phosphorylations (Figure 1B). Notably, we observed consistent protein changes after both inhibitors (AZD2281 and BMN673) in ten cancer cell lines with various backgrounds, indicating its on-target effects and the generalization of the adaptive responses.

In general, the adaptive responses are robust and mainly involved in four aspects (Figure 1B). First, PARPi profoundly upregulated ATM/pATM, pRB, CDK1, CHK1/pCHK1, FOXM1, cyclin B1, CHK2/pCHK2, RAD51, and RAD50, indicating the increased DNA damage and the activation of DNA damage repair pathway, including the activation of the cell cycle DNA damage checkpoints (G1/S, and G2/M phase, whose activation allow time for DNA repair). Second, data also showed the potent activation of the PI3K/AKT/mTOR signaling, including pGSK3-beta, pmTOR, p70S6K, and their downstream targets pS6_pS235_S236, pS6_pS240_S244. Next, we found an unanticipated decrease of FOXO3a, p27, and BIM after PARP inhibition, while there is a modest increase of pBRaf, pMEK, pMAPK, pPKC, and pYB1, indicating the existence of RAS/RAF/MAPK pathway activation. Furthermore, PARPi induced a major imbalance of apoptotic mediators after

PARPis, with upregulation of BAX, BCL-XL, and BID and a remarkable decrease in BIM. As BIM is downstream of both the PI3K and RAS/MAPK pathways, this is to be expected. Most of these pathway disturbance after PARPi have been validated *in vitro* or *in vivo* models in our and others previous works[16,19–21], which will be illustrated in detail below.

Synergistic anti-tumor effects of combination with PARP and PI3K inhibitors

First, as shown in figure 1B, there is a strong evidence for PI3K-AKT-mTOR pathway activation (upregulation of GSK3-alpha-beta_pS21_S9, Rb_pS807_S811, mTOR_pS2448, p70S6K_pT389, and their downstream S6_pS240_S244, S6_pS235_S236) after PARPi. In support of the concept that targeting adaptive rewired pathway would be effective, we and others have demonstrated synergistic interactions between PI3K-AKT-mTOR, and PARP inhibition with a diverse set of inhibitors in multiple preclinical models[22–25]. Furthermore, this has been confirmed in the clinical setting with the PI3K inhibitor (buparlisib) and PARPi (olaparib) in late-stage breast cancers or high-grade serous ovarian cancer (HGSOC), with an ORR of 28% in breast cancer patients, and of 29% in HGSOC regardless of platinum sensitivity (NCT01623349)[23]. Subsequently, concurrent treatment with olaparib and another PI3Kα-specific inhibitor (alpelisib) showed that 36% of ovarian cancer patients, even those BRCA wild-type (somatic and germline) and platinum-resistant ovarian carcinomas, achieved a partial response and 50% ovarian cancer patients had stable disease. Notably, in this clinical trial, 93% (26/28) patients were platinum-resistant, a notoriously therapy-resistant population. Importantly, of the patients with stable disease, around 30% of patients had a durable response for at least six months[24]. Mechanistically, PI3K/mTOR inhibitions were proposed to sensitize cancer cells to PARPi by inducing HRD through repression of genes involved in DDR (BRCA1, RAD51). Taken together, these studies demonstrated that PARPi-based combinations predicted from the CART platform could be validated in preclinical and clinical studies.

Sequential PARP and WEE1 inhibition ameliorate toxicity while maintaining efficacy

As indicated in Figure 1B, the CART showed that PARPi induces DNA damage and replication stress leading to activation of DNA damage responses (ATM, ATM_pS1981) including the S and G2 cell cycle checkpoints (Chk1, Chk1_pS345, Chk2, Chk2_pT68, CDK1, Cyclin_B1, and FoxM1) (Figure 1B). PARP inhibitors remarkably induce DNA damage and subsequent dependency on S-phase and G2/M checkpoint activities for survival. WEE1 is a critical kinase that regulates both S-phase and G2/M phase cell-cycle checkpoints by inhibiting the CDK2 and CDK1, respectively. Thus, we tested the efficacy of the combination with PARPi and Wee1 inhibitors (WEE1i) *in vitro* and *in vivo*. Consistent with previous studies that showed the combination of olaparib and WEE1i demonstrated improved antitumor efficacy in preclinical chemotherapy-refractory small cell lung cancer models[15,26,27], concurrent combination of PARPi and WEE1i was highly effective in ovarian cancer cells and PDX models regardless of HRD status. However, prolonged PARPi and WEE1i therapy were poorly tolerated in mice resulting in weight loss[15]. Again, using RPPA

we revealed that both the PARP and Wee1 inhibitors induced DNA damage and replication stress. More importantly, these effects were sustained for at least several days after drugs removal. Given that, we explored the new sequential therapy strategy with PARP and Wee1 inhibitors in various preclinical models. As expected, the sequential treatment with PARP and Wee1 inhibitors maintained efficacy but significantly ameliorated toxicity[15]. Mechanically, the high basal DNA replication stress in cancer contributed to the selective anti-tumor effect of sequential therapy on tumor cells. Notable, although normal cells are also minimally affected by the drug combination, but since their basal level of replication stress is lower, the sequential treatment is not as toxic as in cancer cells. A clinical trial entitled Sequential Trial of Agents Against DNA Repair (STAR), to assess the therapeutic index of sequential administration of PARPi and WEE1i will be launched soon (NCT04197713).

Moreover, we also found that PARPi induced a remarkable activation of RAS/MAPK pathway, with the profound upregulation of MAPK_pT202_Y204, MEK1_pS217_S221, and BRaf_pS445. In addition, FOXO3a, p27, and BIM were down-regulated (Figure 2A). Further, there was an obvious imbalance of apoptosis after PAPRi with upregulation of BAX, BCL-XL, BID but remarkable decrease in BIM (Figure 1B, 2A). BIM is well-known to be downregulated by the MAPK pathway. So, based on these observations, we assessed the anti-tumor efficacy of concurrent combination of PARPi with MEKi in a panel of ovarian cancer cell lines. Remarkably, PARPi and MEK inhibition demonstrate synergy in a subset of ovarian cancer cell lines and in vivo models, especially in KRAS mutant tumors[16]. Moreover, the RAS/MAPK pathway activation also contribute to the acquired PARPi resistance. Combination of MEK and PARP inhibition reversed acquired PARPi resistance. Mechanically, mutant RAS is associated with higher HR capacity, and MEKi decreases expression levels of multiple components of the HR pathway in RAS mutant cells, aggravating PARP inhibitor-induced DNA damage. In addition to the effects on DNA damage repair, MEKi induced FOXO3a-BIM cascade-mediated apoptosis that contributed to the synergy with PARP inhibitors (Figure 2A–2B). Although mutations of RAS/MAPK pathway are rare in HGSOC, pathway activation is common. Furthermore, KRAS mutation is high in low-grade serous ovarian cancers as well as in endometrial carcinomas. Supported by these preclinical findings[16], we have initiated a clinical trial entitled SOLAR, testing the combination of MEK inhibitor (selumetinib) and olaparib in endometrial, ovarian, and other solid tumors with RAS pathway alterations, as well as those with difficult to treat PARPi resistant ovarian cancers (NCT03162627). Preliminary results from this dose-escalation phase 1 trial recently demonstrated a promising patient's benefit. Given RAS mutations are among the most common drivers of tumorigenesis, and are frequently highly resistant to therapy, the combination of PARPi and MEKi could be of great importance across a broad set of cancer types.

CART in a window of opportunity trial (WOO) of PARPi

The aforementioned CART strategy for PARPi were developed primarily in preclinical models. In order to optimize their development in clinically relevant settings, we further applied CART platforms to identify informative adaptive responses after talazoparib monotherapy in a WOO trial after talazoparib monotherapy in HGSOC[18]. In this WOO trial,

multiple biopsies from different peritoneal lesions were acquired both before drug treatment (during an initial laparoscopy), and after treatment with talazoparib for 7 to 14 days. Collected samples were analyzed by RPPA[18]. Importantly, we found that the 1) CART platform was also applicable for clinical samples. 2) adaptive responses in patients that are also found in cell lines. This support the use of pre-clinical model to study adaptive responses and test combination therapies. 3) although individual patient displays a distinct adaptive response to 1–2 weeks of treatment of talazoparib, there are no obvious interlesional heterogeneity. 4) adaptive responses in patients can also be targeted with drugs (PI3K pathway, G2-M, etc.). Thus, these data from clinical patients further support that the CART platforms could provide an opportunity for patient-specific selection of rational combination therapies designed to abrogate patient-specific adaptive responses to maximize patient benefit.

Conclusion

The CART platform represents a powerful tool to develop rational combination therapy. We have identified a number of rational combination therapies based on adaptive responses, which were validated in preclinical models and subsequently moved to the clinic. In the future, as the number of certified antibodies increases and analysis methods improves, the power of CART will provide opportunities to improved patient outcomes.

Acknowledgements

Funding

GBM is supported by NIH/NCI grants CA217685, CA217842, SAC110052 from Komen Breast Cancer Foundation, the Ovarian Cancer Research Foundation, The Breast Cancer Research Foundation, and a kind gift from the Miriam and Sheldon Adelson Medical Research Fund. ML is supported by the Ovarian Cancer Research Alliance and Ruth and Steve Anderson, in honor of Shae Anderson Gerlinger.

Reference

1. Pilie PG, Tang C, Mills GB & Yap TA State-of-the-art strategies for targeting the DNA damage response in cancer. Nature reviews. Clinical oncology 16, 81–104, doi:10.1038/s41571-018-0114-z (2019).
2. Lord CJ & Ashworth A PARP inhibitors: Synthetic lethality in the clinic. Science (New York, N.Y.) 355, 1152–1158, doi:10.1126/science.aam7344 (2017).
3. Grimaldi G & Corda D ADP-ribosylation and intracellular traffic: an emerging role for PARP enzymes. Biochem Soc Trans 47, 357–370, doi:10.1042/BST20180416 (2019). [PubMed: 30710058]
4. Farmer H et al. Targeting the DNA repair defect in BRCA mutant cells as a therapeutic strategy. Nature 434, 917–921, doi:10.1038/nature03445 (2005). [PubMed: 15829967]
5. Bryant HE et al. Specific killing of BRCA2-deficient tumours with inhibitors of poly(ADP-ribose) polymerase. Nature 434, 913–917, doi:10.1038/nature03443 (2005). [PubMed: 15829966]
6. Wang S et al. Uncoupling of PARP1 trapping and inhibition using selective PARP1 degradation. Nat Chem Biol 15, 1223–1231, doi:10.1038/s41589-019-0379-2 (2019). [PubMed: 31659317]
7. Pommier Y, O'Connor MJ & de Bono J Laying a trap to kill cancer cells: PARP inhibitors and their mechanisms of action. Science translational medicine 8, 362ps317, doi:10.1126/scitranslmed.aaf9246 (2016).

8. Mateo J et al. A decade of clinical development of PARP inhibitors in perspective. Annals of oncology : official journal of the European Society for Medical Oncology 30, 1437–1447, doi:10.1093/annonc/mdz192 (2019). [PubMed: 31218365]
9. Sharma P, Hu-Lieskovan S, Wargo JA & Ribas A Primary, Adaptive, and Acquired Resistance to Cancer Immunotherapy. Cell 168, 707–723, doi:10.1016/j.cell.2017.01.017 (2017). [PubMed: 28187290]
10. Ramos P & Bentires-Alj M Mechanism-based cancer therapy: resistance to therapy, therapy for resistance. Oncogene 34, 3617–3626, doi:10.1038/onc.2014.314 (2015). [PubMed: 25263438]
11. Akbani R et al. A pan-cancer proteomic perspective on The Cancer Genome Atlas. Nat Commun 5, 3887, doi:10.1038/ncomms4887 (2014). [PubMed: 24871328]
12. Li J et al. Characterization of Human Cancer Cell Lines by Reverse-phase Protein Arrays. Cancer Cell 31, 225–239, doi:10.1016/j.ccell.2017.01.005 (2017). [PubMed: 28196595]
13. Vogel C & Marcotte EM Insights into the regulation of protein abundance from proteomic and transcriptomic analyses. Nat Rev Genet 13, 227–232, doi:10.1038/nrg3185 (2012). [PubMed: 22411467]
14. Zhang H et al. Integrated Proteogenomic Characterization of Human High-Grade Serous Ovarian Cancer. Cell 166, 755–765, doi:10.1016/j.cell.2016.05.069 (2016). [PubMed: 27372738]
15. Fang Y et al. Sequential Therapy with PARP and WEE1 Inhibitors Minimizes Toxicity while Maintaining Efficacy. Cancer Cell 35, 851–867 e857, doi:10.1016/j.ccell.2019.05.001 (2019). [PubMed: 31185210]
16. Sun C et al. Rational combination therapy with PARP and MEK inhibitors capitalizes on therapeutic liabilities in RAS mutant cancers. Science translational medicine 9, doi:10.1126/scitranslmed.aal5148 (2017).
17. Sun C et al. BRD4 Inhibition Is Synthetic Lethal with PARP Inhibitors through the Induction of Homologous Recombination Deficiency. Cancer Cell 33, 401–416 e408, doi:10.1016/j.ccell.2018.01.019 (2018). [PubMed: 29533782]
18. Labrie M et al. Adaptive responses in a PARP inhibitor window of opportunity trial illustrate limited functional interlesional heterogeneity and potential combination therapy options. Oncotarget 10, 3533–3546, doi:10.18632/oncotarget.26947 (2019). [PubMed: 31191824]
19. Sand A et al. WEE1 inhibitor, AZD1775, overcomes trastuzumab resistance by targeting cancer stem-like properties in HER2-positive breast cancer. Cancer Lett 472, 119–131, doi:10.1016/j.canlet.2019.12.023 (2020). [PubMed: 31866466]
20. McGrail DJ et al. Proteome Instability Is a Therapeutic Vulnerability in Mismatch Repair-Deficient Cancer. Cancer Cell, doi:10.1016/j.ccell.2020.01.011 (2020).
21. Sun C et al. BRD4 Inhibition Is Synthetic Lethal with PARP Inhibitors through the Induction of Homologous Recombination Deficiency. Cancer Cell 33, 401–416.e408, doi:10.1016/j.ccell.2018.01.019 (2018). [PubMed: 29533782]
22. Ibrahim YH et al. PI3K inhibition impairs BRCA1/2 expression and sensitizes BRCA-proficient triple-negative breast cancer to PARP inhibition. Cancer Discov 2, 1036–1047, doi:10.1158/2159-8290.CD-11-0348 (2012). [PubMed: 22915752]
23. Matulonis UA et al. Phase I dose escalation study of the PI3kinase pathway inhibitor BKM120 and the oral poly (ADP ribose) polymerase (PARP) inhibitor olaparib for the treatment of high-grade serous ovarian and breast cancer. Annals of oncology : official journal of the European Society for Medical Oncology 28, 512–518, doi:10.1093/annonc/mdw672 (2017). [PubMed: 27993796]
24. Konstantinopoulos PA et al. Olaparib and alpha-specific PI3K inhibitor alpelisib for patients with epithelial ovarian cancer: a dose-escalation and dose-expansion phase 1b trial. The Lancet. Oncology 20, 570–580, doi:10.1016/S1470-2045(18)30905-7 (2019). [PubMed: 30880072]
25. Juvekar A et al. Combining a PI3K inhibitor with a PARP inhibitor provides an effective therapy for BRCA1-related breast cancer. Cancer Discov 2, 1048–1063, doi:10.1158/2159-8290.CD-11-0336 (2012). [PubMed: 22915751]
26. Karnak D et al. Combined inhibition of Wee1 and PARP1/2 for radiosensitization in pancreatic cancer. Clinical cancer research : an official journal of the American Association for Cancer Research 20, 5085–5096, doi:10.1158/1078-0432.CCR-14-1038 (2014). [PubMed: 25117293]

27. Lallo A et al. The Combination of the PARP Inhibitor Olaparib and the WEE1 Inhibitor AZD1775 as a New Therapeutic Option for Small Cell Lung Cancer. Clinical cancer research : an official journal of the American Association for Cancer Research 24, 5153–5164, doi:10.1158/1078-0432.Ccr-17-2805 (2018). [PubMed: 29941481]

Perspectives

- Adaptive responses is a key mechanism of PARPi resistance, but also represent therapeutic vulnerabilities that can be targeted through combination therapies.
- The reverse phase protein array (RPPA) technology is a powerful, high-throughput, quantitative, cost-effective technology for functional proteomics studies of adaptive responses
- Combinatorial Adaptive Response Therapy (CART) platform is a powerful tool to develop rational combination therapy capitalizing on adaptive responses

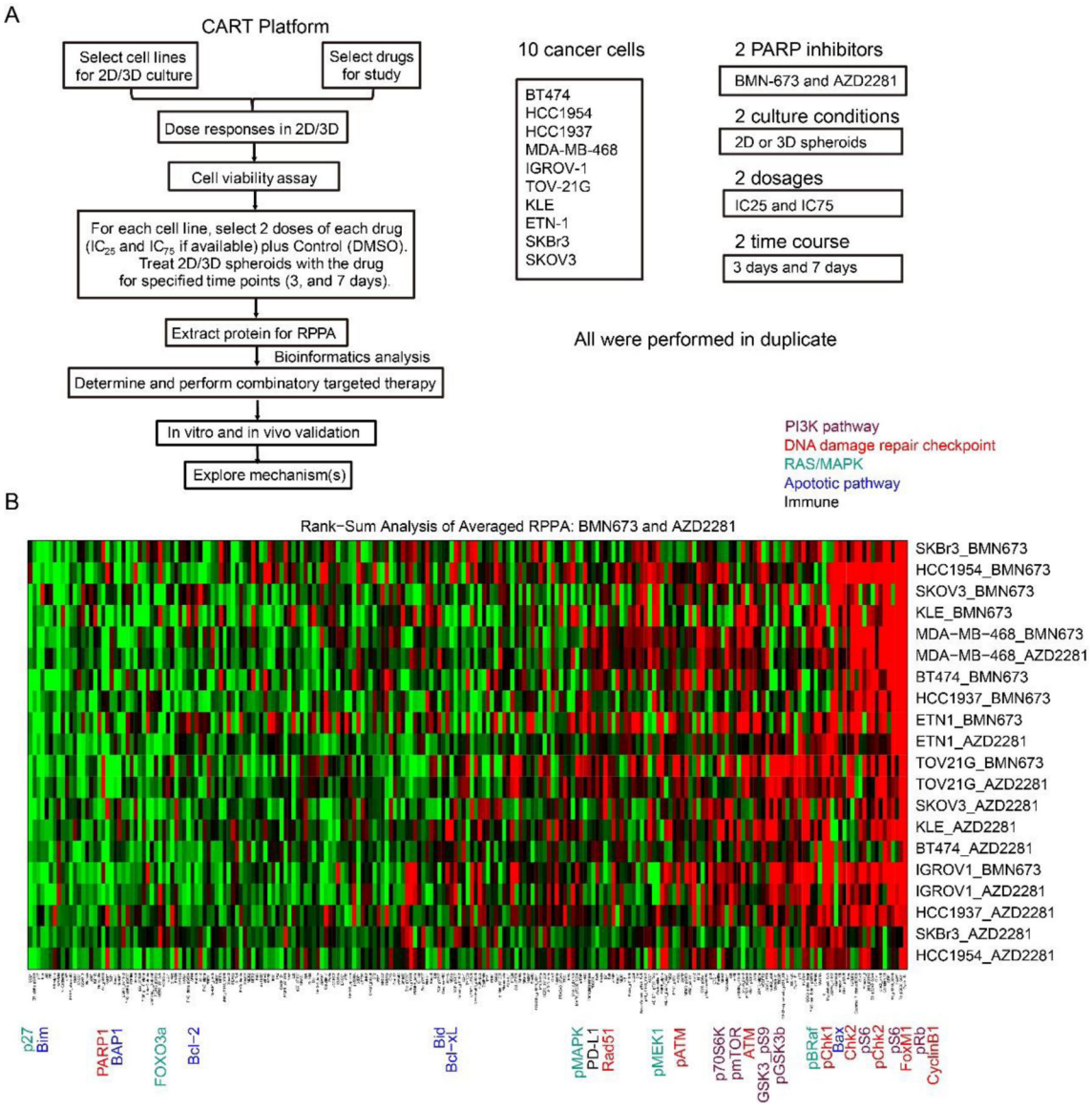

Figure 1. Rational combination therapies development based on adaptive response to PARP inhibitors evaluated by RPPA

(A) Flow chart of CART platform after PARP inhibitors.

(B) Ten cell lines were treated as indicated in the flow chart (A), then cell lysates were evaluated by RPPA with 218 antibodies targeting total and phosphorylated proteins and phosphorylation. Heatmap shows the integrated proteins or phosphorylation alterations following both AZD2281, and BMN673 treatment. To display the generalization, proteins amounts in various treatment conditions (2D/3D, time-points, and dosage) were averaged. Protein levels were normalized to each individual control for AZD2281, or BMN673 respectively, and then ordered by the summing median-centered numbers.

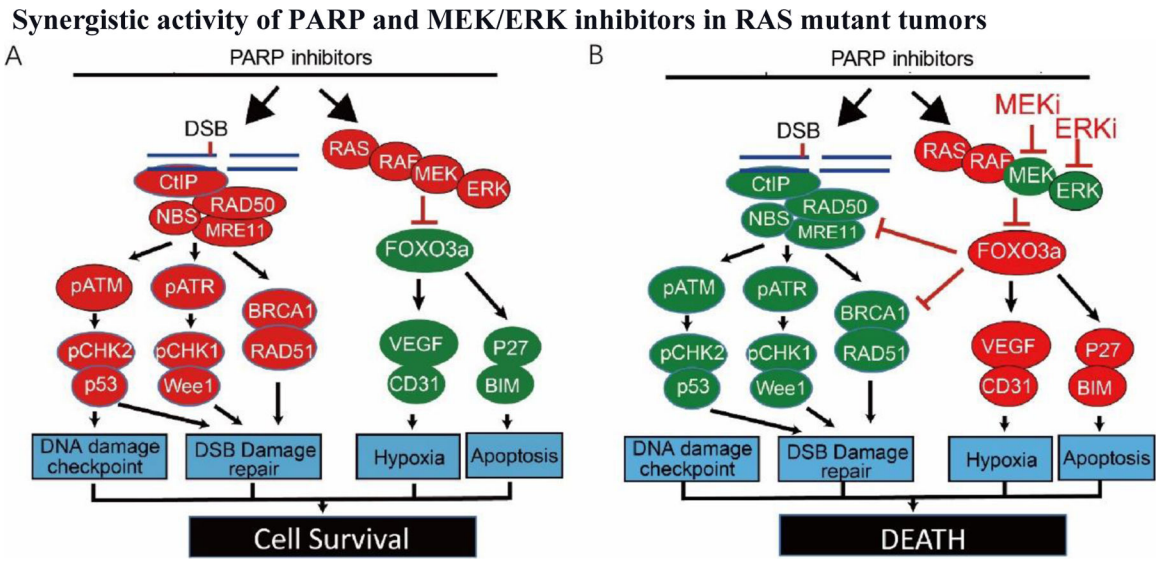

Figure 2. Concurrent MEK inhibition reverses the activated MAPK pathway after PARPi and leads to cell to death.

(A) PARP inhibition induced a marked increase in RAS/mitogen-activated protein kinase (MAPK) pathway activation, (B) Concurrent MEK inhibition reverses the adaptive responses after PARPi and lead cell to death.