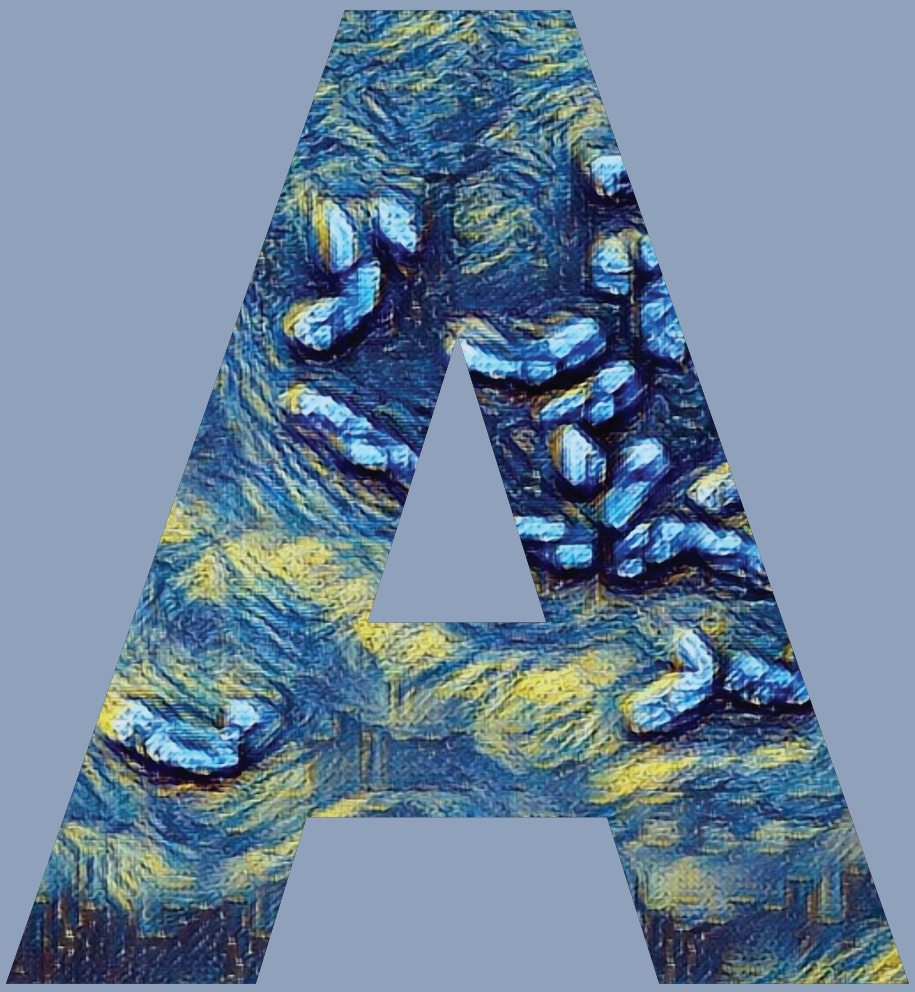
New dimensions of the cellular response to DNA damage

Moser, S.C.



APPENDICES

ENGLISH SUMMARY

Thirty years ago, scientists discovered that mutations in the *BRCA1* gene strongly predispose women to develop breast and ovarian cancers. This was a landmark finding as it provided the first direct evidence that not only environmental factors, but also inherited changes in our DNA, cause cancer. Soon after, researchers found that BRCA1 plays a key role in repairing damaged DNA, in particular double-stranded DNA breaks (DSBs) through a process called homologous recombination (HR). HR uses undamaged DNA as a template to repair the break without errors to prevent the loss of genetic information. If women inherit a mutated *BRCA1* gene, this repair is impaired. As a result, their cells accumulate DNA mistakes (mutations) over time, putting them at a significantly elevated risk of developing cancer.

In **Chapter 2**, we discuss our current understanding of BRCA1 biology, how BRCA1-related cancer is initiated, how patients with *BRCA1*-mutant tumors are currently treated, and ongoing efforts to prevent BRCA1-associated cancer from developing in the first place.

During my PhD, I investigated the mechanism of action of HR and explored how two frequently used treatment options, platinum-based chemotherapy and PARP inhibitors (PARPi), kill HR-deficient tumors. I also explored how cancers become unresponsive to these treatments and searched for alternative ways to target resistant cancers.

HR is not performed by BRCA1 alone but requires many other proteins, one of them being RAD51, which helps to find the correct DNA sequence to use as a template for repair. Surprisingly, little is known about what happens after RAD51 finds the matching sequence. In **Chapter 3**, we performed genetic screening experiments in human cells and identified a new HR factor: the previously uncharacterized protein C1orf112/FIRRM. FIRRM helps to remove RAD51 from the damaged site once its job is finished and recruits other proteins to complete repair of the DNA break. Cells without FIRRM are extremely sensitive to chemotherapy and PARPi. Mice that lack FIRRM die before birth, and deleting FIRRM in breast tissue of mice accelerates breast cancer development. These findings reveal FIRRM as an important new factor required for repairing DSBs via HR and suggest that testing FIRRM levels may predict which patient population will respond best to platinum-based chemotherapy and PARPi.

Chapter 4 delves deeper into platinum-based chemotherapy (like cisplatin), which works by creating DNA damage that cells with dysfunctional HR are unable to repair. Using tumor samples from patients with breast cancer that were transplanted and grown in mice (patient-derived xenograft models), we found that analyzing a tumor's genome and the DNA scars that remain when repair fails can help predict a tumor's response to platinum agents. We also identified mutations in two genes, *XRCC3* and *ORC1*, that sensitize tumor cells to platinum-based chemotherapy, similar to *BRCA1* mutations.

Chapters 5 and **6** focus on PARPi, a newer class of drugs that exploit a concept called "synthetic lethality". While cells can survive with either a *BRCA1* mutation or under PARPi treatment alone, combining both is lethal to cancer cells, but spares healthy cells. This makes PARPi attractive because they specifically target *BRCA1*-mutant cancer cells. In **Chapter 5**, we summarize the current clinical use of PARPi, how tumors get resistant to treatment and strategies to overcome it using drug combinations.

In **Chapter 6**, we explored novel avenues to target PARPi-resistant tumors and made an unexpected discovery: PARPi do not only damage DNA but also disrupt DNA packaging. They cause histones, proteins that wrap and protect DNA, to be temporarily removed. A group of helping proteins, including the INO80 complex, NASP and PARP1 remove these histones, store them and put them back once the DNA break is repaired. Importantly, when disturbing this process, by removing NASP, cancer cells become very sensitive to PARPi. This so-called "histone supply chain" could therefore be a promising target for treating drug-resistant tumors.

Finally, **Chapter 7** discusses the discoveries made during my PhD research, connects them and points towards future challenges: mapping the final steps of HR in greater detail, understanding how DNA packaging affects treatment response and designing smarter drug combinations that exploit a tumor's unique weaknesses. Taken together, this work advances our understanding of how cells repair DNA and proposes new strategies to outsmart tumors, bringing us closer to more effective, personalized cancer treatments for affected individuals.