Laboratory of Cancer Cell Biology

Cell cycle, checkpoint, DNA damage response, phosphorylation, oncogenic transformation

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Proliferation of cells is essential for keeping organisms alive and healthy and is accomplished by passing through interphase followed by nuclear division (mitosis) and cellular division (cytokinesis). In response to DNA damage, cells temporarily stop progression through the cell cycle (checkpoint) to prevent transmission of mutations to progeny. After completion of DNA repair, cells are allowed to re-enter the cell cycle (checkpoint recovery). Cells that are exposed to a massive genotoxic stress that exceeds the capacity of DNA repair are eliminated by programmed cell death. Radiotherapy and chemotherapy with genotoxic pharmaceuticals represent two commonly used non-surgical strategies in treatment of human tumours and they both rely on induction of cell death by genotoxic stress. Progression through the cell cycle and cellular responses to DNA damage are tightly controlled by interconnected signalling cascades. Malfunction of cellular checkpoints causes accumulation of mutations and can lead to genome instability, activation of oncogenes, and eventually to malignant transformation.

In our recently established laboratory we employ cell biology, molecular biology and biochemical approaches to identify the molecular mechanisms that control cellular responses to DNA damage. In particular, we focus on protein phosphatase PPM1D/Wip1 that plays an essential role in switching off the DNA damage response pathway, termination of the checkpoint and control of checkpoint recovery. PPM1D/Wip1 is an important negative regulator of the tumour suppressor p53. Recent data from transgenic mice and from human tumours implicate PPM1D/Wip1 as an oncogene. Our work aims to decipher the molecular mechanisms regulating the function of PPM1D/Wip1 in human cells and in mouse models. In addition, we use chemical genetics to evaluate PPM1D/Wip1 as a potential pharmacological target.

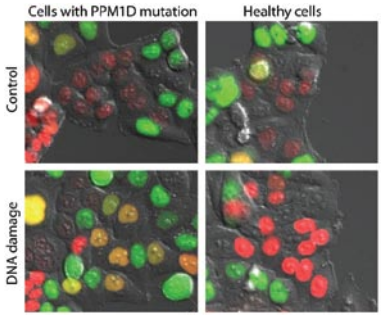

Fig. 1. Healthy cells do not proliferate in the presence of DNA damage and stop progression through the cell cycle in G1 phase (red). Cells carrying mutation in exon 6 of the PPM1D gene continue cell division despite the presence of DNA damage, progress to S phase and replicate their DNA (orange). Mutation in PPM1D does not affect cells in G2 (green). Courtesy of Indra A. Shaltiel and Libor Macurek

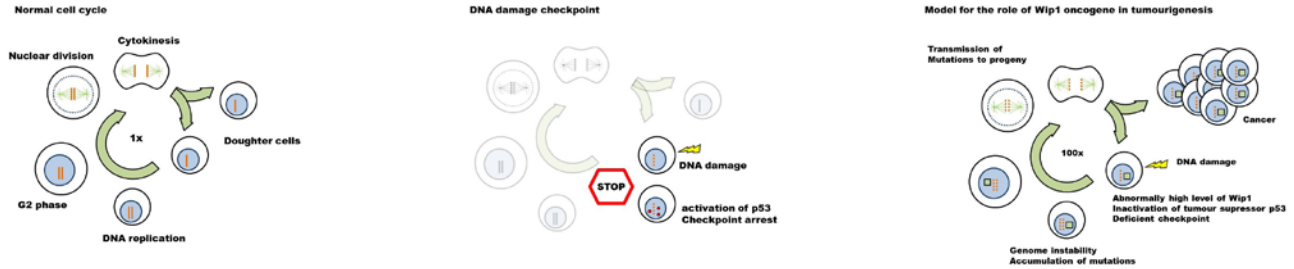

Fig. 2. Model for PPM1D mutation in cancer

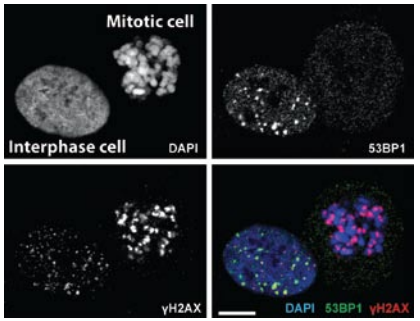

Fig. 3. After exposure to DNA damage, interphase cells form foci positive for 53BP1 and gH2AX. Mitotic cells form only gH2AX-positive foci but fail to recruit 53BP1 to the sites of DNA damage.

- GACR, GAP305/12/2485 – Structure and function of proteins involved in DNA damage signalling, 2012-2015, L. Macůrek
- GACR, GA13-18392S – Dynamics of DNA damage response in cells, 2013-2016, L. Macůrek
- GACR, 14-34264S – Role of R2TP complex in DNA damage response and cell proliferation, 2014-2016, Z. Líčeníková Hořejší
- MEYS, 7F14061 – Phosphorylation-mediated signalling in DNA damage response and cancer, 2014-2017, L. Macůrek
- Worldwide Cancer Research – Role of PPM1D/Wip1 truncating mutations in cancer predisposition, 2014, L. Macůrek

1. Benada J, Burdová K, Lidak T, von Morgen P, Macurek L: Polo-like kinase 1 inhibits DNA damage response during mitosis. **Cell Cycle** (in press).
2. Bruinsma W, Macurek L, Freire R, Lindqvist A, Medema RH: Bora and Aurora-A continue to activate Plk1 in mitosis. **J Cell Sci 2014** 127(Pt 4): 801-11.
3. Kleiblova P, Shaltiel IA, Benada J, Ševčík J, Pecháčková S, Pohlreich P, Voest EE, Dundr P, Bartek J, Kleibl Z, Medema RH, Macurek L: Gain-of-function mutations of PPM1D/Wip1 impair the p53-dependent G1 checkpoint. **J Cell Biol 2013** 201(4): 511-21.
4. Macurek L, Benada J, Müllers E, Halim VA, Krejčíková K, Burdová K, Pecháčková S, Hodný Z, Lindqvist A, Medema RH, Bartek J: Downregulation of Wip1 phosphatase modulates the cellular threshold of DNA damage signaling in mitosis. **Cell Cycle 2013** 12(2): 251-62.
5. Sevcik J, Falk M, Macurek L, Kleiblova P, Lhota F, Hojny J, Stefancikova L, Janatova M, Bartek J, Stribrna J, Hodny Z, Jezkova L, Pohlreich P, Kleibl Z: Expression of human BRCA1•17-19 alternative splicing variant with a truncated BRCT domain in MCF-7 cells results in impaired assembly of DNA repair complexes and aberrant DNA damage response. **Cell Signal 2013** 25(5): 1186-93.

From the left: Tomáš Liďák / Diploma Student, Soňa Pecháčková, MSc / PhD student, Jan Benada, MSc / PhD student, Andra Stefania Vieru, MSc/ PhD student (since October 2014), Libor Macůrek, MD, PhD / Head of Laboratory, Kamila Burdová, PhD / Postdoctoral fellow (since December 2014), Gabriela Jeníková, PhD (part time) / Postdoctoral fellow, Patrick von Morgen, MSc / PhD student (since October 2014), Monika Burócziová, PhD (part time) / Postdoctoral fellow

Not in the picture: Zuzana Líčeníková Hořejší, PhD / Research Fellow (since January 2014), Petra Kleiblová, MD, PhD (part time) / Postdoctoral fellow