Identification of CDK9 as a New Interacting Partner of BRCA1, BARD1 and PTIP Tandem BRCT Domains.

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INTRODUCTION Eukaryotic cells are constantly subjected to endogenous and exogenous DNA damage agents. In response to this genotoxic stress they evolved a DNA damage response (DDR) pathway. This response consists in an elaborated protein-protein interaction pathway that regulates other cellular processes as cell cycle control, apoptosis and the DNA damage repair itself. Several proteins involved in the DDR present the BRCT domain, which can exist as a single or tandem domain (tBRCT), which is capable of binding phosphorylated peptides. Recently, our group described a protein interaction network that identified putative interacting partners of seven different proteins enclosing the tBRCT. Among other hits we identified Cyclin-Dependent Kinase 9 (CDK9) as a common partner of BRCA1, BARD1 and PTIP tBRCTs. CDK9 is a component of the positive transcription elongation factor- b complex (P-TEFb), which is involved in the transcriptional elongation, co-transcriptional histone modification, mRNA processing and mRNA export. Recently CDK9 was shown to function directly in genome integrity maintenance in response to replication stress.

MATERIAL AND METHODS In order to confirm the CDK9 interaction with the BARD1 we used protein-protein interaction routines such as GST pulldown and immunoprecipitation assays. We are extending our approach in order to confirm the CDK9 protein interactions with BRCA1 and PAXIP.

RESULTS AND DISCUSSION We were able to confirm and map the BARD1 interaction with the two isoforms of CDK9 (42KDa and 55KDa). We also observed the interaction between an ectopically expressed 55KDa CDK9 and constitutive BARD1 in HEK293FT cells.

CONCLUSION The characterization of these new CDK9 interactions will help to unravel the role of this kinase in the DDR and genome stability maintenance pathways.

Palavra chave: CDK9, Câncer, tBRCT
Patrocínio: FAPESP, IFRJ, INCA, CNPq and CAPES