Instituto de Medicina Molecular João Lobo Antunes

Master Project Proposal

Title: Identification and study of novel ATRS alleviators in Alternative Lengthening of Telomeres

Synopsis:

Telomeres are nucleoprotein structures that avoid that the ends of linear eukaryotic chromosomes are recognized as DNA double-stranded breaks (DSBs). They are comprised of 5'-TTAGGG-3' tandem repeats, the multiprotein complex shelterin and the long noncoding RNA TERRA, an RNA Polymerase II (RNA Pol II) transcript produced from telomeres. Because telomeres naturally shorten at each cell division, most cancer cells activate specialized machineries to buffer telomere shortening and thus avert replicative senescence. The most common mechanism of telomere elongation in cancers relies on telomerase activity; however, about 15% of cancers are telomerase-negative and elongate their telomeres through homology-directed repair mechanisms referred to as Alternative Lengthening of Telomeres (ALT). Based on this conservative estimate, almost 1 million people per year die of ALT-positive cancers worldwide. ALT is prevalent in brain and bone cancers and ALT cancers are extremely aggressive and resilient to current therapeutic strategies.

ALT relies on Break-Induced Replication (BIR)-mediated elongation of replicatively stressed telomeres. We have recently proposed that this ALT-specific Telomeric Replication Stress (ATRS) must be kept at physiological levels sufficient to induce BIR, yet not high enough to interfere with cell proliferation. This balance is achieved by the counteraction of molecular triggers and alleviators of ATRS, whose nature is only now starting to be understood. In agreement, we have recently reported that the Fanconi Anemia Complementation group M (FANCM) translocase is a bona fide ATRS alleviator. When FANCM is depleted in ALT cells, ATRS is exacerbated, cells stop proliferating and rapidly die. These effects are not observed in non-cancerous cells nor in ALT-negative cancer cells that maintain their telomeres through the action of telomerase. Based on these observations, we proposed that ATRS alleviators represent extremely promising targets for novel therapies for ALT cancers.

Based on the rationale that alleviators of ATRS should bind more avidly to replicatively stressed ALT telomeres, we have performed a proteomics study to identify factors that are enriched at those telomeres. This project will combine molecular and cellular biology and high-end microscopy to confirm if the identified factors are novel ATRS alleviators essential for ALT cell survival and depict their molecular functions at ALT telomeres.

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Bibliography:

Arora R, Lee YW, Wischnewski H, Brun CM, Schwarz T, and Azzalin CM. (2014) RNaseH1 regulates TERRA-telomeric DNA hybrids and telomere maintenance in ALT tumour cells. Nat Commun *5*:5220.

Silva B, Pentz R, Figueira AM, Arora R, Lee YW, Hodson C, Wischnewski H, Deans AJ, and Azzalin CM. (2019). FANCM limits ALT activity by restricting telomeric replication stress induced by deregulated BLM and R-loops. Nat Commun *10*:2253.

Domingues-Silva B, Silva B, and Azzalin CM. (2019). ALTernative Functions for Human FANCM at Telomeres. Front Mol Biosci *6*:84.