Internal ribosome entry site (IRES)-mediated protein synthesis in colorectal cancer progression

Plano de trabalho:

The regulation of protein synthesis plays a critical role in mediating cell growth. Translation initiation is tightly regulated by a balanced concentration of factors involved in protein synthesis. In the majority of cases (canonical translation) translation initiation is brought about by the cap-dependent mechanism. However, several oncogenes, growth factors and proteins involved in the regulation of programmed cell death contain a different translation initiation mechanism mediated by internal ribosome entry sites (IRESs). IRES-dependent translation requires several trans-acting proteins known as IRES trans-acting factors (ITAFs). This mechanism involves the direct recruitment of the ribosome to the vicinity of the initiation codon without the involvement of the cap structure. This alternative to the cap-dependent mechanism of translation initiation can sustain protein synthesis under stress conditions and promote tumorigenesis.

Colorectal cancer (CRC) represents one of the leading causes of cancer mortality in the Western world. Here we propose to identify proteins which are synthesized by a non-canonical translation initiation mechanism when colorectal tumour cells face metabolic stress. For this, canonical translation initiation will be impaired by RNA interference (RNAi) against canonical initiation factors, in different cell lines subjected to several stress stimuli, including nutrient deprivation and hypoxia. Proteins with altered expression levels will be identified by mass spectrometry. The corresponding mRNAs will be studied to identify IRESs. We plan to characterize the IRES functional mechanisms and corresponding ITAFs. Moreover, we will validate the physiological significance of these selected IRES-mediated proteins in CRC tumorigenesis. We truly believe that the expected results will prompt us for the better understanding of a further mechanism contributing to the survival of cancer cells, or to therapy resistance.

Metodologias a utilizar:
Cultura de células e transfecção; clonagem; *RNA interference*; extração de DNA e RNA; sequenciação; RT-PCR; RT-qPCR; Western blot; imunoprecipitações; tradução *in vitro*; ensaios de bioluminescência; etc.

Vagas: 1 estudante

Duração: 1 ano lectivo

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