“Investigating the function of the ribosomal protein S24 long isoform on cellular survival during hypoxic stress”

Historically, ribosomal subunits were thought to only have a constitutive role in translation and were viewed as passively active. Recently, it has been discovered that ribosomes can be heterogenous and selectively translate specific mRNAs. In response to cellular stress, such as hypoxia (low oxygen), eukaryotic cells have developed expression control mechanisms at the translational level in order to survive during stress. Given that hypoxia is a characteristic of the tumour microenvironment, hypoxic stress response mechanisms provide tumour cells with survival benefits, thereby increasing oncogenic potential. One mechanism used to combat hypoxia is alteration of ribosomal protein stoichiometry within the ribosome in order to preferentially translate transcripts that elicit a stress response. Recent studies in the Uniacke lab analyzed alternative splice variants of ribosomal protein S24 (RPS24) and found that the long isoform increased in spheroids (in vitro aggregates of tumour cells) relative to the short isoform. Therefore, I hypothesize that the RPS24 long isoform provides the ribosome with a specialized function increasing survival within the hypoxic spheroid microenvironment. First, I will compare the stability of the RPS24 long and short isoforms using a cycloheximide chase assay to determine if the relative increase of the RPS24 long isoform can be attributed an increase in stability. Next, I will use polsosome profiling to determine if there is a difference in the translatome of ribosomes containing the RPS24 long isoform versus the short isoform. Finally, I will evaluate the effects of RPS24 long and short protein isoforms on tumour cell viability in vitro using live/dead staining. This research will aid our understanding of alternative mechanisms used by the cell during hypoxic stress.