Chapter 1

Introduction

1.1 Background

Pancreatic Cancer (PaCa) is the tenth most common cancer in the UK with approximately 10,500 new cases every year and the incidence rate is suggested to increase by 5% between 2023-2040 (Cancer Research UK, 2023). According to Surveillance, Epidemiology, and End Result (SEER) Database (2023), PaCa is associated with a terminal prognosis with 1-year survival rates of 11.5% and 5-year survival rates below 5%. Consequently, it remains the fifth leading cause of cancer-related death with 9,000 death cases in the UK yearly (Pancreatic Cancer UK, 2023). The poor prognosis of PaCa is correlated with chemoresistance and desmoplasia as the major contributors. Desmoplasia is a fibroinflammatory response resulting in reduced elasticity of the tumour tissue with increase in interstitial fluid pressure (IFP). The chronic inflammation that characterizes desmoplasia involves infiltration of immune cells such as neutrophils, tumour-associated macrophages (TAM), and regulatory T cells (Treg). The complex signalling arising from the infiltration creates an immunosuppressive microenvironment that promotes PaCa growth, spread and therapeutic resistance (Whatcott et al, 2012).

Adrenomedullin (AM) is a hormone that has been shown to promote progression of various tumours (Vazques et al, 2021). AM mediates the polarization of tumour-associated macrophages in Melanoma, promoting tumour growth and angiogenesis *in vitro* and *in vivo* (Chen et al, 2011). AM also induces mast cell degradation, therefore, promoting proliferation and inhibiting apoptosis of Gastric Cancer *in vitro* and *in vivo* (Lv et al, 2018). AM binds to Adrenomedullin Receptor (AMR) which formed by the combination of Class B G-Protein Coupled Receptors (GPCR) called Calcitonin Receptor-Like Receptor (CLR) with the Receptor Activity-Modifying Proteins (RAMPs). The interaction of GPCR with RAMP1 and RAMP2 give rise to Adrenomedullin-1 Receptor (AM1R) and Adrenomedullin-2 Receptor (AM2R), respectively (Hay & Pioszak, 2016).

GPCRs are the largest family of cell surface receptors. Increasing evidence show GPCR signalling is an important mechanism in PaCa development, and therefore, GPCRs represent the largest class of druggable targets for cancers, especially PaCa. The role of RAMPs in the GPCR family adds an extra complexity to this drug class. RAMPs are a family of single transmembrane domain proteins GPCR. Recent study showed that small molecules were designed to target the AM signalling through inhibition of AM2R, without affecting the signalling via AM1R, were efficacious at decreasing PaCa growth *in vitro* and *in vivo* (Avgoustou et al, 2020). However, since the role of RAMPs are important in the modulation of receptor signalling, RAMPs also shown to be interacted with other class of GPCR. Therefore, study the individual RAMPs is also important rather than focusing on the CLR/RAMP complexes. Thus, this study was conducted to analyse the effect of individual RAMPs towards cancer cells viability and apoptosis, with focus to validate the effect of RAMPs deficiency by knockdown the RAMPs using siRNA.

1.2 Hypothesis

RAMPs signalling affects pro-tumoral characteristics in a panel of PaCa cells in vitro.

1.3 Objectives

The main objectives of this research are to understand the wider implications of the RAMP family in PaCa and to identify the effect of RAMP signalling towards the pro-tumoral characteristics of PaCa cells in-vitro.

Other than that, the effect of RAMP1 and 2-mediated signalling to PaCa and the inflammatory stimuli effect to the expression of RAMP1 and 2 were investigated.