Seema Hijazi
Undergraduate
LSU Health Sciences Center, New Orleans, LA

Mentors: Rinku Majumder, PhD, and Samarpan Majumder, PhD
LSUHSC, Department of Interdisciplinary Oncology and Department of Genetics

"Protein S Modulates the Hypoxic Response in Pancreatic Cancer Cells"

Objective: Pancreatic ductal adenocarcinoma (PDAC) is the third leading cause of cancer-related mortality in the United States and remains one of the deadliest malignancies due to its aggressive biology, late-stage diagnosis, and limited therapeutic options. Tumor hypoxia is a defining feature of PDAC and promotes metabolic reprogramming, tumor progression, immune evasion, and resistance to therapy. Patients with PDAC also have a markedly increased risk of venous thromboembolism (VTE), which is frequently associated with reduced circulating levels of Protein S (PS), a vitamin K-dependent anticoagulant. While Protein S is well recognized for its anticoagulant function, accumulating evidence from our laboratory suggests that hypoxia suppresses PS expression, raising the possibility that PS also modulates hypoxia-driven signaling pathways. In this study, we investigated whether exogenous Protein S attenuates hypoxia-induced signaling in human pancreatic cancer cells.

Method: Human pancreatic cancer cell lines PANC-1 and MIA PaCa-2 were treated with 150 nM recombinant human Protein S and cultured under normoxic or hypoxic conditions for 8, 24, and 48 hours. Protein expression was analyzed by Western blotting to determine the levels of hypoxia-inducible factor-1α (HIF-1α), hypoxia-inducible factor-2α (HIF-2α), phosphorylated AKT (p-AKT), total AKT, and glucose transporter-1 (GLUT1), using glyceraldehyde-3-phosphate dehydrogenase (GAPDH) as the loading control. Quantitative real-time PCR (RT-qPCR) was performed to assess the expression of hypoxia-responsive target genes.

Results: Exposure to hypoxia markedly increased HIF-1α and HIF-2α protein expression in both PANC-1 and MIA PaCa-2 cells, with the most robust and reproducible induction observed after 8 hours of hypoxic exposure. Treatment with Protein S significantly attenuated hypoxia-induced accumulation of both HIF-1α and HIF-2α in each cell line compared with hypoxia alone, suggesting that Protein S suppresses hypoxia-responsive signaling.
Hypoxia also increased AKT phosphorylation, consistent with activation of pro-survival signaling pathways. In contrast, total AKT and GLUT1 protein levels remained largely unchanged under the experimental conditions. Ongoing RT-qPCR analyses of hypoxia-responsive genes will determine whether the reduction in HIF-1α and HIF-2α protein levels is accompanied by corresponding decreases in the transcriptional activation of downstream target genes.

Conclusion: These findings demonstrate that Protein S suppresses hypoxia-induced accumulation of HIF-1α and HIF-2α in human pancreatic cancer cells, supporting a previously unrecognized role for Protein S as a negative regulator of the cellular hypoxic response. Collectively, these preliminary data suggest that Protein S modulates hypoxia-driven signaling in PDAC and provide a strong mechanistic rationale for future studies to define its molecular mechanisms of action and evaluate its therapeutic potential as a novel target for pancreatic cancer.