"A Novel Role of the Anticoagulant Protein S in Preventing Thrombosis"

Blood coagulation is an intricate process that occurs by the actions of numerous procoagulants and anticoagulants. Protein S (PS) is a vitamin K-dependent anticoagulant whose physiological importance is underscored by several hematological disorders in PS-deficient individuals. Genetic deficiency of PS is associated with increased risk of venous thrombosis and recurrent thrombosis, e.g., familial venous thrombosis. Acquired PS deficiency predisposes to (recurrent) venous thromboembolism and loss of fetus. PS knockout mice are embryonic lethal. Currently, three disparate functions of PS are known: 1) PS is a cofactor for activated protein C, 2) PS is a cofactor of tissue factor pathway inhibitor, and 3) PS is a direct inhibitor of coagulation Factor IXa.

Deep vein thrombosis, cardiovascular diseases, and stroke are common among obese individuals, as well as among individuals who have high body mass index. Obesity is a major health threat to the global population. According to 2014 statistics, 13% of the world’s population is obese, and more than 42% of the USA population is obese. The liver is a major organ that is negatively affected by obesity. For instance, obesity causes diseases like nonalcoholic fatty liver disease. In obesity-induced diseases, the liver develops hypoxia. Hypoxia stabilizes transcription factors such as Hypoxia Inducible Factor-1α (HIF-1α) by preventing Von Hippel-Lindau-mediated hydroxylation of HIF-1α and its degradation. Recently, work from Dr. Majumder’s laboratory showed that an increase in HIF-1α expression downregulates Protein S expression in HEP-G2 cells. The goal of this project was to determine whether hypoxia associated with obesity results in a prothrombotic state because of downregulation of PS.

We measured thrombin generation (a prothrombotic state indicator) with a thrombin generation assay, and free PS concentration was measured by ELISA assays. We used plasma from HIF1α knockout mice and the HIF1α P564A mutant (HIF1α dPA) mice which are resistant to degradation, resulting in sustained, elevated HIF1 protein abundance, even under normal O2 concentrations. We observed a higher amount of thrombin in obese and HIF1α dPA mice compared with the control mice. However, HIF1α knockout mice generated less thrombin, like the control mice. Additionally, we observed that the clotting times of HIF1α dPA mice was significantly higher than in HIF1α KO mice. This result showed the effect of HIF1α on PS expression and thrombin generation. Because the mice were exposed to more hypoxic conditions, they were more likely to have a decreased PS level.

Completion of these studies will provide understanding of the regulation of PS expression. This project will be instrumental in investigating new antithrombotic strategies for chronic complications that have high thrombotic risk, such as obesity.