CALIFORNIA STATE SCIENCE FAIR
2008 PROJECT SUMMARY

Name(s)

Paul Tran

Project Number

S0419

Project Title

The Inhibitory Effect of HKa on Prostate Cancer Line DU145 Is Mediated by Blocking Metastasis

Abstract

Objectives/Goals

Prostate Cancer is defined by the uncontrollable proliferation of prostate cells. When growth factors bind to their receptors, a transactivation signal is sent to EGFR and uPAR, resulting in clustering of EGFR and uPAR. This increases Cyclin D1 expression and facilitates migration of prostate cancer cells. High-molecular weight kininogen [HK] is a plasma protein responsible blood-clotting. Cleavage of HK results in the release of bradykinin and cleaved high molecular weight kininogen [HKa]. HKa induces apoptosis of proliferating epithelial cells and inhibits angiogenesis in vivo. The purpose of this experiment was to prevent the metastasis of prostate cancer cells through implementation of HKa. HKa would bind to uPAR and prevent the transactivation signaling from growth factors to EGFR. This competitive inhibition would inhibit metastasis of prostate cancer cells.

Methods/Materials

Sub-cultured Prostate Caner DU145 cells into six 35mm2 dishes coated with 1mL Collagen [10¥ìg/mL] until monolayer is formed. Measured metastasis/migration of Prostate Cancer DU145 cells in vitro through a Migration Assay. 3 dishes were Control. 3 dishes were HKa Treated. Added 6.09¥ìL HKa [100nM] to HKa Treated dishes. Added 2mL Pure Media [DMEM + L-Glut] with Zn2+ [15¥ìM] and 2¥ìL bFGF [20ng/mL] to each dish. Incubated each dish at 37¢ªC for 24 hours. After incubation, fixed tumor cells with 2mL Formalin Solution [1.2mL DPBS Buffer + 0.8mL 10% Formaldehyde] and incubated at 4¢ªC. Pictures of each migration line were taken at 0 and 24 hours to show contrast/growth of cells in the Control and HKa Treated dishes.

Results

EGFR and uPAR co-localized on the surface of Prostate Cancer DU145 cells in response to bFGF [20ng/ml]. Immunofluorescence microscopy showed that HKa disrupted co-localization of EGFR and uPAR, indicating that HKa prevented transactivation signaling from growth factors to EGFR. HKa inhibited migration of DU145 cells at 24 hours by the Migration Assay. A monoclonal antibody against uPAR also blocked migration of DU145 cells at 24 hours by the Migration Assay. It suggested that HKa inhibited migration of tumor cells by targeting uPAR.

Conclusions/Discussion

HKa disrupted the interaction of EGFR and uPAR and inhibited migration of Prostate Cancer DU145 cells across migration line. It indicates that HKa might prevent metastasis of human Prostate Cancer.

Summary Statement

My project investigates the use of cleaved high molecular-weight kininogen, a plasma protein, in inhibiting the metastasis of Prostate Cancer Cell Line DU145 in vitro.

Help Received

Participant in Physician Scientist Training Program; gained mentorship/workspace from faculty at Sol Sherry Thrombosis Research Center at Temple University School of Medicine in Philadelphia, PA.