Targeting GDPD5 and GDPD6 in Orthotopic Human Breast Cancer Xenograft Models: A Metabolomics study

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Synopsis

Activated choline phospholipid metabolism is a hallmark of cancer. Aggressive breast cancers are characterized by high tumoral phosphocholine and glycerophosphocholine. In our ongoing efforts of evaluating the glycerophosphodiesterases GDPD5 and GDPD6 as cancer treatment targets, we have systemically injected mice growing orthotopic triple-negative MDA-MB-231 breast tumors with lentiviral vectors that silence the GDPD5 or GDPD6 genes as compared to mice injected with control viruses. We have analyzed extracted tumor tissue by means of high-resolution 1H MRS-based metabolomics. Differences in tumor growth and metabolic profiles were observed following silencing of GDPD5 and GDPD6 genes when compared to control mice.

Introduction

- Breast cancer is the second most common cancer and cause of death in women in the United States.[1]
- Previous studies have shown that glycerophosphocholine (GPC), although elevated as compared to normal tissue, is lower than phosphocholine (PC) in aggressive breast cancer.[2]
- Two enzymes that are responsible for cancer-related alteration in GPC have recently been identified: The glycerophosphocholine-phosphodiesterases (GPC-PDEs) GDPD5 and GDPD6.[3-4]
- These enzymes cleave GPC to glycerol-3-phosphate and free choline, and are overexpressed in breast cancer.
- The in vitro GDPD5 and GDPD6 transient silencing[5] studies on different breast cancer cell lines showed significant increase in GPC level.[6]
- We have continued our studies in vivo to evaluate if shRNA-expressing lentiviral vector based silencing of GDPD5 and GDPD6 will reduce tumor growth in orthotopic MDA-MB-231 breast cancer xenograft model.
- We have studied the effects of silencing these genes using high resolution 1H Magnetic Resonance Spectroscopy at the metabolic level in our continuing efforts of identifying potential biomarkers as well as therapeutic targets for anti-cancer therapy.

Materials and Methods

- Prior to lentiviral treatment, tumors showed a gradual increase in size. Upon lentiviral treatment, tumor growth was significantly reduced in the GDPD6-shRNA treated group (Figure 1A)
- Representative bright-field and fluorescence micrographs showing GFP distribution in treated tumor tissue sections are presented (Figure 1B)
- High-resolution 1H MR spectra of the water phases of tumor extract of the three treated groups demonstrates differences in various metabolites (Figure 2 and 3)
- A heatmap of all relevant metabolites was generated based on percent (%) changes among the three groups
- Significant changes were observed in metabolites constituting various metabolic pathways such as amino acid metabolic pathway, post-translational and glycolytic pathway etc. which are in accordance with literature.

Results and Discussion

- Levels of choline, GPC and PC also varied among the groups, signifying the altered choline phospholipid metabolism.
- Choline was decreased whereas GPC was increased in GDPD6-shRNA treated group (Figure 5A)
- The PC/GPC ratio was lower in the GDPD6-shRNA treated group as compared to groups treated with luc-shRNA and GDPD5-shRNA (Figure 5B)
- The results showed the expected outcome of GDPD6 knockdown.

Conclusions

- Our results suggest that GDPD5 and GDPD6 silencing in breast cancer tumor xenografts could be an effective molecular treatment strategy.
- The detected metabolites showed varying degrees of occurrence and response to the treatment with GDPD5-shRNA and GDPD6-shRNA treatment.
- Additional studies are continuing these initial findings.

Literature Citations


Further Information

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