

CALIFORNIA STATE SCIENCE FAIR 2008 PROJECT SUMMARY

Name(s)

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Project Number

S0401

Project Title

The Study of Tamm-Horsfall Protein in Interstitial Cystitis

Objectives/Goals

Abstract

Tamm-Horsfall protein (THP) is the most abundant urinary glycoprotein that has a protective function in the bladder mucous layer. THP has anionic properties due to the presence of a negatively charged sugar called sialic acid, which plays an important role in the THP#s protective function. It has been shown that the total content of sialic acid has been reduced in patients with Interstitial Cystitis (IC) compared to normal. This project is set out to determine whether the aggregation property of THP is different in IC patients compared to normal because of reduced sialic acid content, and thus could be used as a simple diagnostic tool.

Methods/Materials

THP was isolated from the urine samples of 16 control and 16 IC patients by the salt precipitation method. The samples were analyzed for their total sialic acid content by the DMB-HPLC assay. First, sialic acid was released by mild acid hydrolysis, and then tagged with DMB. The sialic acid was quantified using RP-HPLC with C18 column and fluorescent detection. For the aggregation study, the purified THP was dissolved in a phosphate buffer containing 4M urea. Samples were then analyzed by size exclusion chromatography using HPLC with Superdex-200 column and UV detection.

Results

The DMB-HPLC assay indicated that the sialic acid content was two-times higher in normal compared to IC patients. The size exclusion chromatography using Superdex-200 of THP showed a major THP peak at 7.5 minutes in both normal and IC patients. However, there was a second peak at 17 minutes, which was present in 75% of IC patients and 19% of normals.

Conclusions/Discussion

Interstitial cystitis (IC) is a chronic bladder disorder in which there is a defect in the protective function of the bladder epithelium. In patients with IC, urinary solutes such as potassium penetrate the bladder epithelium and provoke symptoms of pain, urgency, and frequency. IC is often misdiagnosed because its symptoms are similar to other diseases. Size exclusion chromatography of THP could be used to differentiate THP of IC patients from normal because of the presence of a second peak. Since this method does not need any derivatization, it can be used as a simple and fast diagnostic tool to identify IC.

Summary Statement

The study of aggregation properties of THP by size exclusion chromatography can be used as a diagnostic tool because it showed a difference between the IC patient versus normal.

Help Received

Dr. C. Lowell Parsons allowed me to carry out the experiments in his lab at UCSD; Marianne Chenoweth obtained the urine samples; Dr. Sulabha Argade supervised me in the lab.