

技术与方法

大鼠附睾液蛋白质组二维液相色谱分离的实验研究

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摘要 目的: 建立一种利用二维液相色谱分离和质谱鉴定大鼠附睾液蛋白质组的实验方法, 为以后研究其它哺乳动物附睾液蛋白质组提供实验基础。方法: 分离提取大鼠附睾头体尾部官腔蛋白, 样品经脱盐、浓缩, 利用起始缓冲液置换, 进行一维色谱聚焦分离, 收集pH8.5-4.0之间的组份进行二维反相色谱分离, 并通过自动馏分收集器收集分离组份; 最后将获得的二维UV图通过ProteoVue软件转换成二维PI/UV图谱; 选择含高丰度蛋白的馏份经冷冻干燥浓缩后进行质谱鉴定。结果: 通过二维液相色谱成功分离了大鼠头体尾部附睾液蛋白并建立了二维PI/UV图谱; 对二维获得的含高丰度蛋白的馏分进行了质谱鉴定, 获得大鼠附睾头体尾部主要蛋白。结论: 为进一步利用二维液相色谱全面分离附睾蛋白和研究附睾功能奠定了基础。

关键词 [蛋白质组](#) [二维液相色谱分离](#) [附睾蛋白质](#)

分类号

Experimental Research on rat epididymal fluid proteins by two-dimensional liquid chromatography

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Abstract **OBJECTIVE:** To establish an experimental method of studying rat epididymal fluid proteome by two dimensional liquid chromatography and MALDI-TOF MS and provide a foundation of studying other mammal epididymal proteome. **METHODS:** The epididymal fluid proteins extracted were exchanged with start buffer and separated by chromatofocusing in the first dimension, the fractions between pH8.5 and pH4.0 collected from 1D were separated by reverse-phase HPLC, the fractions were collected by automatic fraction collector; The UV maps were transformed into PI/UV maps by ProteoVue software; The fractions including high abundance proteins were identified using MALDI-TOF MS. **RESULTS:** We successfully isolated rat epididymal fluid proteins by two dimensional liquid chromatography, constructed PI/UV maps and obtained some high abundant proteins (ERABP, Clusterin, CRISP). **CONCLUSION:** Our studies provide a foundation for further isolating of epididymal fluid proteins and studying of epididymal functions.

Key words [Proteome](#) [Two dimensional liquid chromatographic fractionation](#) [Epididymal proteins](#)

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