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论著

大鼠肾组织中小分子代谢物的GC-MS分析

刘韶[1], 王方杰[1], 梅文娟[2], 陶立坚[2]

1. 中南大学湘雅医院 药剂科, 长沙 410008;
2. 中南大学湘雅医院 肾内科, 长沙 410008

摘要： 目的: 采用气相色谱-质谱法(gas chromatography-mass spectrometry, GC-MS)结合化学计量学分析测定大鼠肾组织中小分子代谢物。方法: 色谱柱为HP-5MS石英毛细管柱(30 m × 0.25 μm × 0.25 mm), 初始柱温为100 ℃, 保持3 min, 程序升温为8 ℃/min至300 ℃, 保持6 min。肾脏组织绞碎后加甲醇萃取, 十七酸为内标, 离心分离得上清液, 氮气吹干, 再用甲氧胺吡啶肟化、衍生化试剂衍生后进行GC-MS分析, 对重叠色谱峰采用化学计量学分辨, 得各化合物的纯色谱与光谱曲线, 通过质谱库检索定性化合物, 同时结合标准品并参考文献确定化合物结构, 用内标法对各成分进行半定量。结果: 共鉴定出53种成分, 肾组织中主要代谢物成分为尿素、氨基酸、糖类与脂肪酸。结论: GC-MS辅以化学计量学法相比单用GC-MS分析, 结果更准确可靠, 将为采用代谢组学技术研究肾脏病变分子标记物提供基础。

关键词： 肾组织 代谢物 气相色谱-质谱 化学计量学

GC-MS determination of metabolites in rat kidneys

LIU Shao[1], WANG Fangjie[1], MEI Wenjuan[2], TAO Lijian[2]

1. Department of Pharmacy, Xiangya Hospital, Central South University, Changsha 410008, China;
2. Department of Kidney, Xiangya Hospital, Central South University, Changsha 410008, China

Abstract: Objective: To establish a method to determine the metabolites in rat kidney tissues by gas chromatography-mass spectrometry (GC-MS) combined with chemometric techniques.
Methods: Metabolites were separated and identified on HP-5MS column (30 m × 0.25 μm × 0.25 mm). The initial column temperature was 100 ℃ lasting 3 min, and then programmed at 8 ℃/min to 300 ℃, maintaining at this temperature for 6 min. The internal standard was heptadecanoic acid. The grinded kidney tissue was exacted by methanol. The supernatant was dried by nitrogen. After the oximation and derivation, the supernatant was analyzed by GC-MS. The overlapped peaks were resolved into pure chromatogram and mass spectra with chemometric techniques. Qualitative analysis was performed by comparing the obtained pure mass spectra with those in NIST mass spectra database and certificated by the standards and the references. The internal method was used for semi-quantitation.
Results: A total of 53 compounds were identified. The main constitutions in the kidney tissue were amino acids, saccharides, fatty acids and urea.
Conclusion: The combination of methods is rapid and accurate for the analysis of metabolites in the kidney tissue, which provides more information for further study of metabonomics in kidney tissues.

Keywords: kidney tissue metabolite GC-MS chemometrics

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通讯作者: 陶立坚,Email:taolj@mail.csu.edu.cn

作者简介: 刘韶,博士,副主任药师,主要从事代谢组学在肾病中应用的研究。

作者Email: taolj@mail.csu.edu.cn

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