



中国科学院水生生物研究所

INSTITUTE OF HYDROBIOLOGY, CHINESE ACADEMY OF SCIENCES

分析测试中心

液相色谱仪与四级杆飞行时间质谱仪 (LC-QTOF) 的原理及应用

门君

2020年7月10日



中国科学院水生生物研究所

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主要内容

- 一、负责平台介绍**
- 二、高效液相色谱仪的原理及应用**
- 三、LC-QTOF的原理及应用**
- 四、平台成员及服务内容**



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一、负责平台介绍

分析测试中心的介绍

成立于2010年



260台套仪器

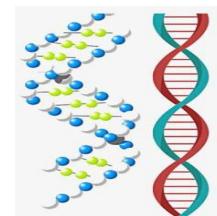
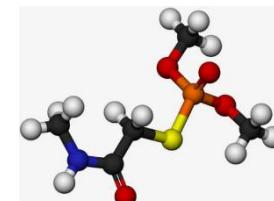
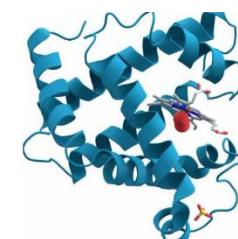


仪器价值1.6亿

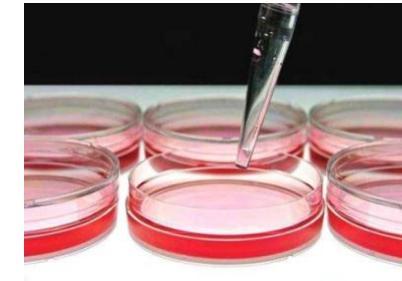


宏观 \longrightarrow 微观

生态 —— 细胞 —— 组织 —— 蛋白 —— 小分子化合物 —— 基因



环境化学技术平台----无机物及元素分析



多功能离子色谱仪

无机阴、阳离子



ICP-OES

Na K Ca Mg Fe Al P
等



ICP-MS

Pb Mn Cu Se As Cr
Cd等

代谢组学技术平台----小分子有机物分析



高效液相色谱仪



全自动氨基酸分析仪



UPLC-MS/MS



LC-QTOF

核苷酸、氨基酸、抗生素、激素、神经递质、
农残等多组分的定量和定性

- 靶向化合物筛查、定性：精确质量数 $< 5\text{ppm}$
- 非靶向代谢组学



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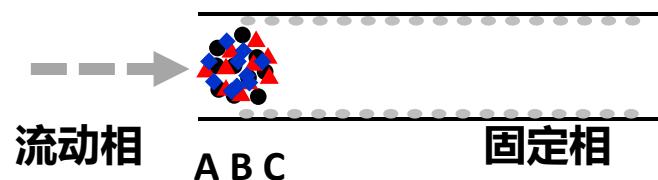


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二、液相色谱仪的原理及应用

色谱法的原理——多组分的分离

- 是利用混合物中各组份在**两相**中溶解、分配、吸附等性能的差异，当两相作相对运动时，使各组分在两相中反复多次受到上述各作用力而达到相互分离。
- 两相中有一相是固定的，叫作**固定相(Stationary Phase)**，有一相是流动的，称为**流动相(Mobile Phase)**。



色谱法的原理——色谱的分类

➤ 按流动相的物态分：

– 气相色谱 (Gas Chromatography, GC)

用气体作为流动相(又叫载气)

适合分离分析**易汽化、热稳定、不易分解**的样品，特别适合
同系物、同分异构体的分离。



– 液相色谱 (Liquid Chromatography, LC)

用液体作为流动相(又叫洗脱剂)

适合分离分析**高沸点、热不稳定、离子型**样品。

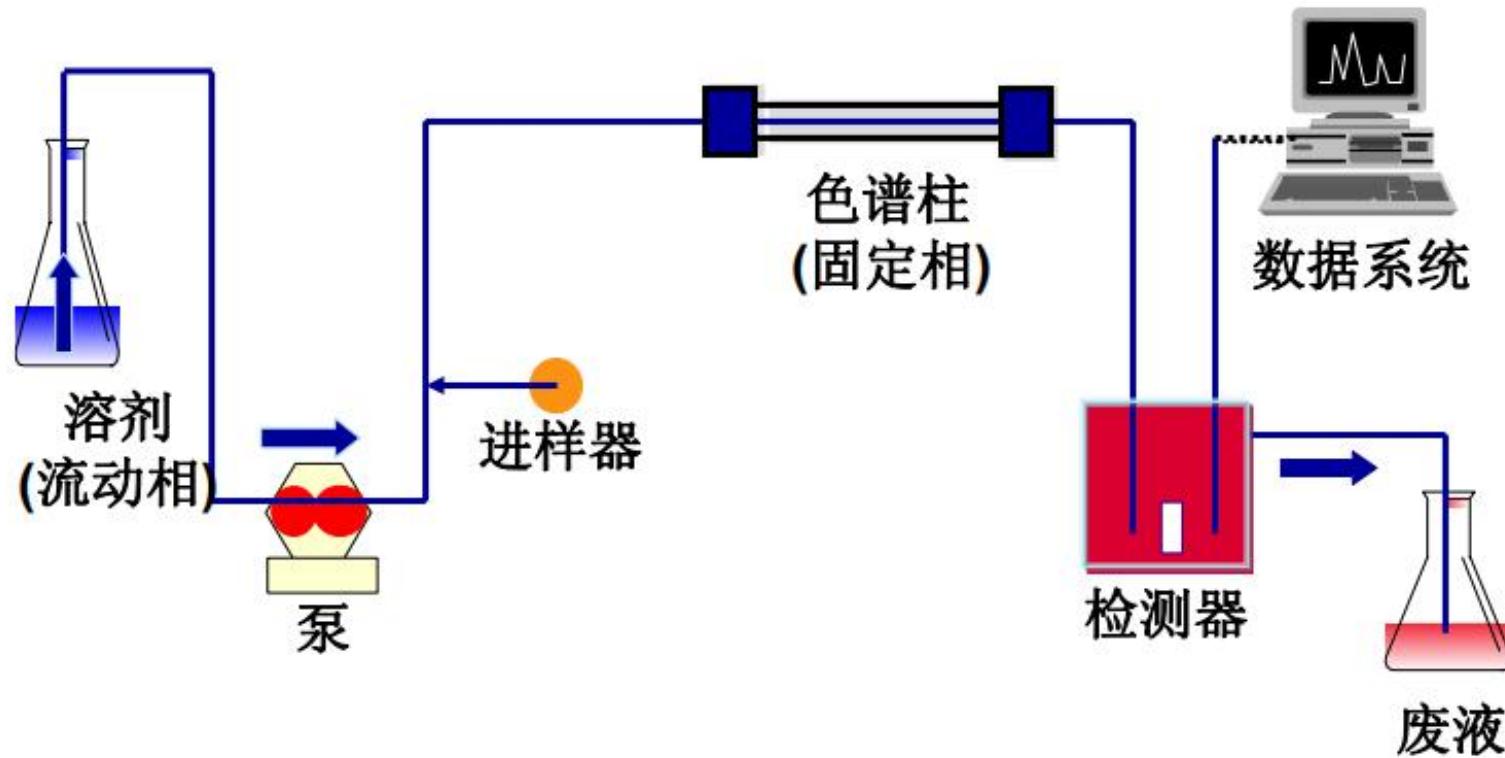


高效液相色谱仪的原理——结构组成

Waters 2695



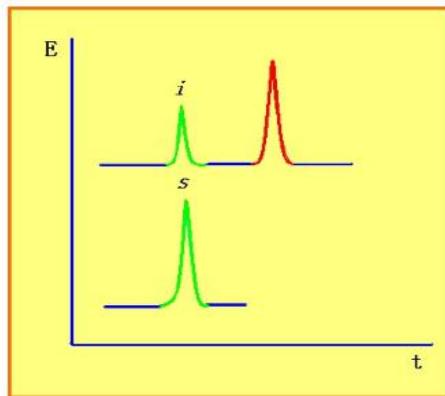
高效液相色谱仪的原理——工作流程



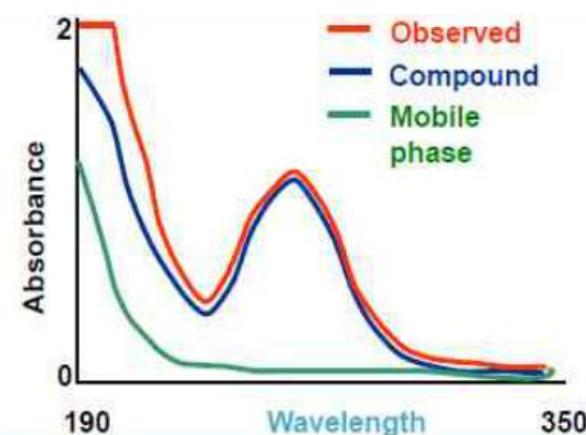
高效液相色谱仪的原理——定性与定量分析

定性分析

(一) 保留时间对照定性



(二) 吸收光谱对照定性



定量分析

(一) 外标法

应用范围：常用于常规分析；

优点：操作简单、计算方便；

(二) 内标法

应用范围：基质干扰大，回收率低；

优点：提高回收率和测量准确性；

高效液相色谱仪的原理——色谱柱及流动性的选择



- 明确方法的目的 (分析 ? 制备 ?)
- 明确分离目的 (单一含量分析 ? 多组分分离 ?)
- 了解样品性质 (极性 ? 酸碱性 ? 分子量大小 ?)
- 查阅相关文献 (相同目标物 ? 相同官能团 ?)

液相检测方法的建立流程



➤ 案例一：鱼肌肉组织中多种呈味核苷酸的测定

检测目的



腺嘌呤核苷酸(腺苷酸，AMP)
鸟嘌呤核苷酸(鸟苷酸，GMP)
胞嘧啶核苷酸(胞苷酸，CMP)
尿嘧啶核苷酸(尿苷酸，UMP)
次黄嘌呤核苷酸(肌苷酸，IMP)

检测物特点

在水溶液和碱溶液中稳定；
酸性($\text{pH} < 4$)溶液中稳定性较差；
加热易发生降解；
极性较强；
有254nm下有较强紫外吸收

➤ 案例一：鱼肌肉组织中多种呈味核苷酸的测定

色谱柱及流动相选择

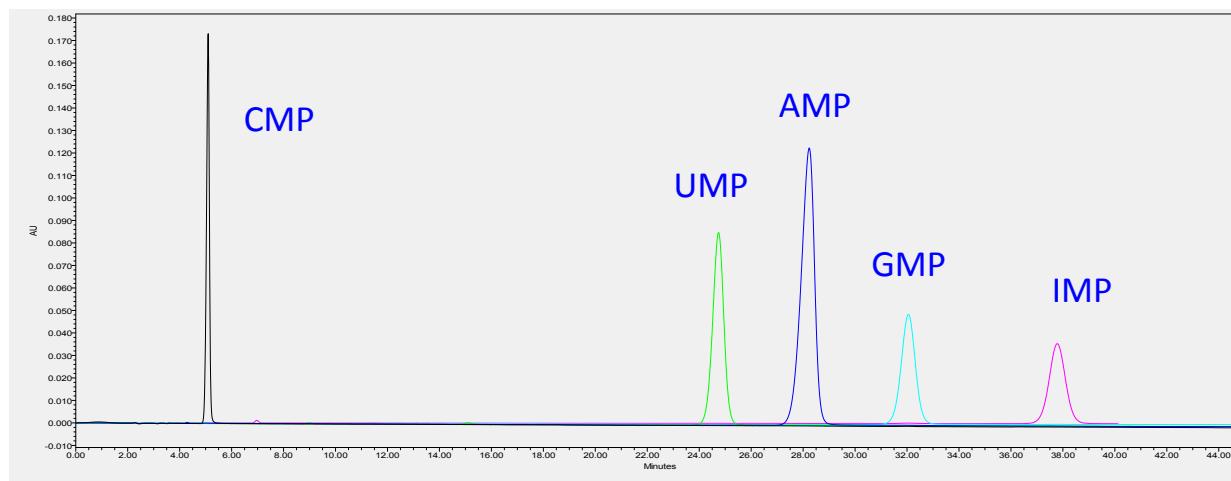


条件： 色谱柱型号：CNW C18-WP 4.6*250mm

流动相： 甲醇 : 磷酸盐缓冲液 (四丁基硫酸氢铵) PH=3.3

流动相比例： 40 : 1000

检测波长： 254nm



标液浓度50mg/L

➤ 案例一：鱼肌肉组织中多种呈味核苷酸的测定

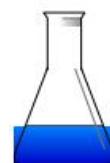
预实验的测定

①样品前处理：

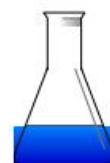


②确定样品中待测物浓度范围、加标回收：

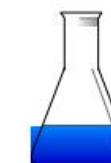
空白组



样品组



加标组

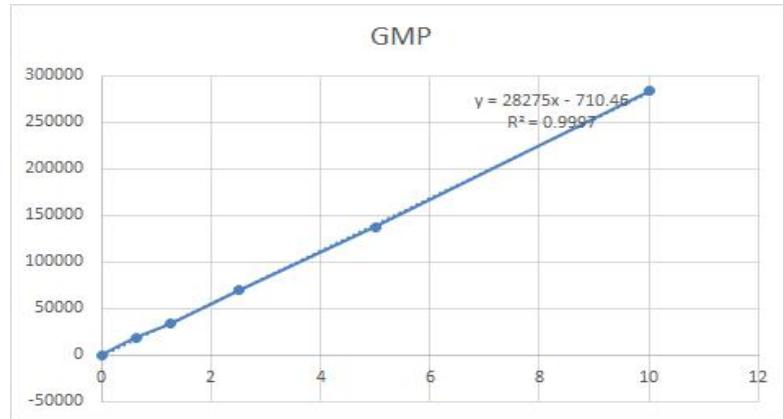


加入已知浓度的标液

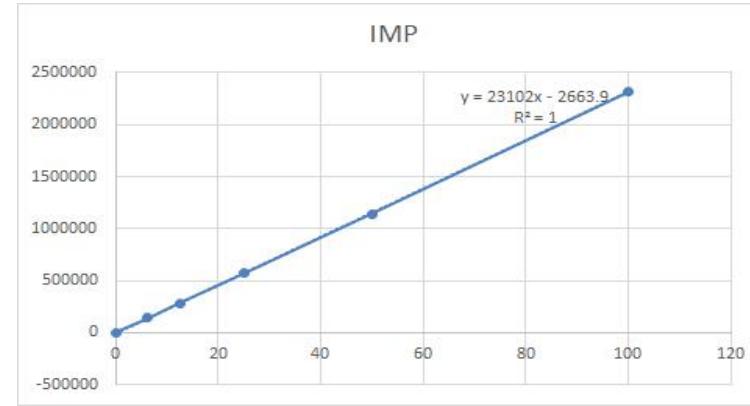


➤ 案例一：鱼肌肉组织中多种呈味核苷酸的测定

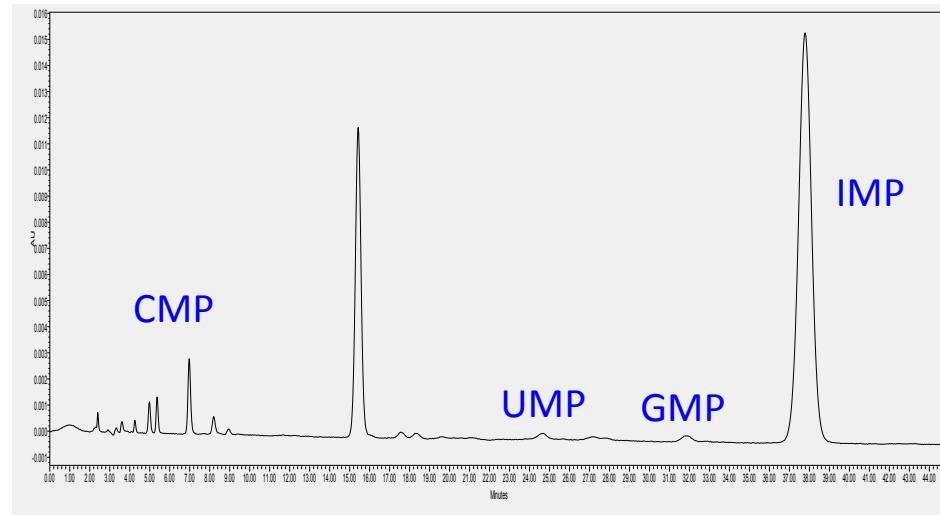
样品的定量检测



GMP线性范围 0-10mg/L



IMP线性范围 0-100mg/L



➤ 案例二：红豆杉中五种紫杉烷类化合物的检测

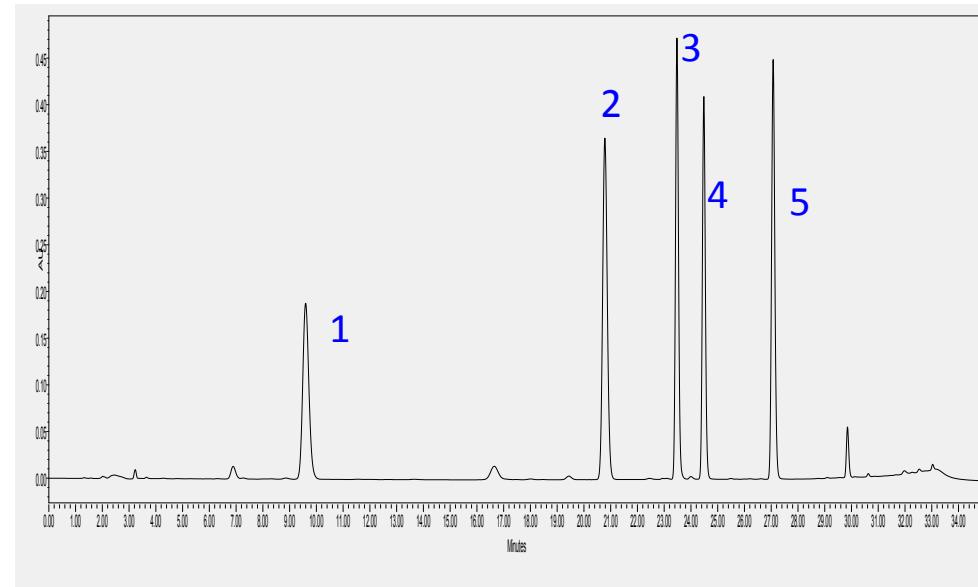
液相检测条件：

流动相： A : 水 B : 乙腈

检测波长： 230nm

流速： 1.0 ml/min

色谱柱型号： Zorbax Eclipse C18 4.6*250mm



序号	化合物
1	10-DABⅢ
2	巴卡亭Ⅲ
3	7-木糖-10-脱乙酰紫杉醇
4	7-表-10-脱乙酰基三尖杉宁碱
5	7-表-10-脱乙酰基紫杉醇

流动相梯度：

Time(min)	0	12	20	30	35	35.1	37	38	40
A(%)	70	70	46	46	10	0	0	70	70
B(%)	30	30	54	54	90	100	100	30	30

➤ 案例三：红球藻中虾青素含量的测定

液相检测条件：

流动相：
A : 二氯甲烷 : 甲醇 : 乙腈 : 超纯水
5 : 85 : 5.5 : 4.5 (v/v)
B : 二氯甲烷 : 甲醇 : 乙腈 : 超纯水
25 : 28 : 42.5 : 4.5 (v/v)

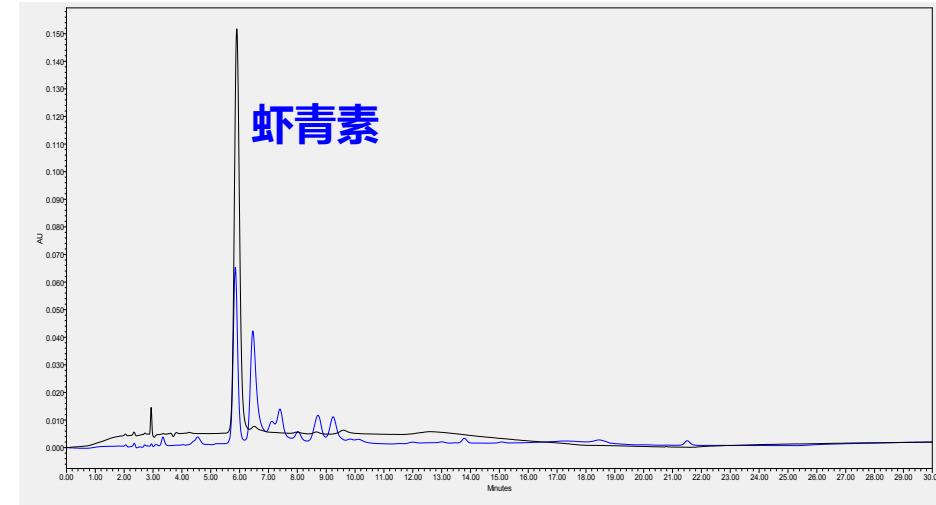
检测波长： 474nm

流速： 1.0ml/min

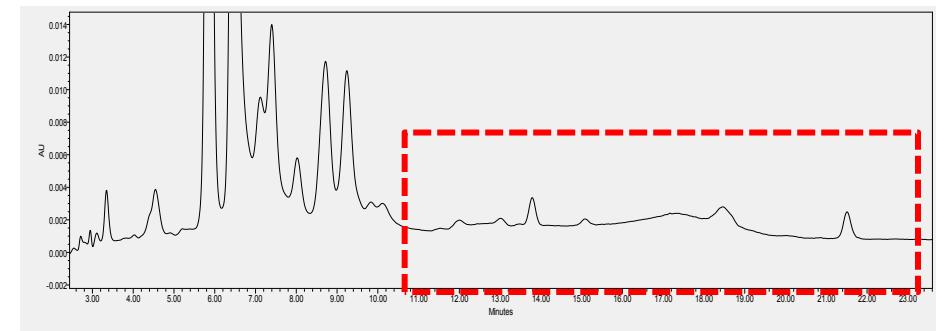
色谱柱型号： Waters C18 4.6*250mm

流动相梯度：

Time(min)	0	8	14	27	28	30
A(%)	100	100	0	0	100	100
B(%)	80	80	100	100	0	0



标液与样品检测图谱比对



样品局部放大图

➤ 案例四：鱼组织中视黄醇等五种化合物的检测

液相检测条件：

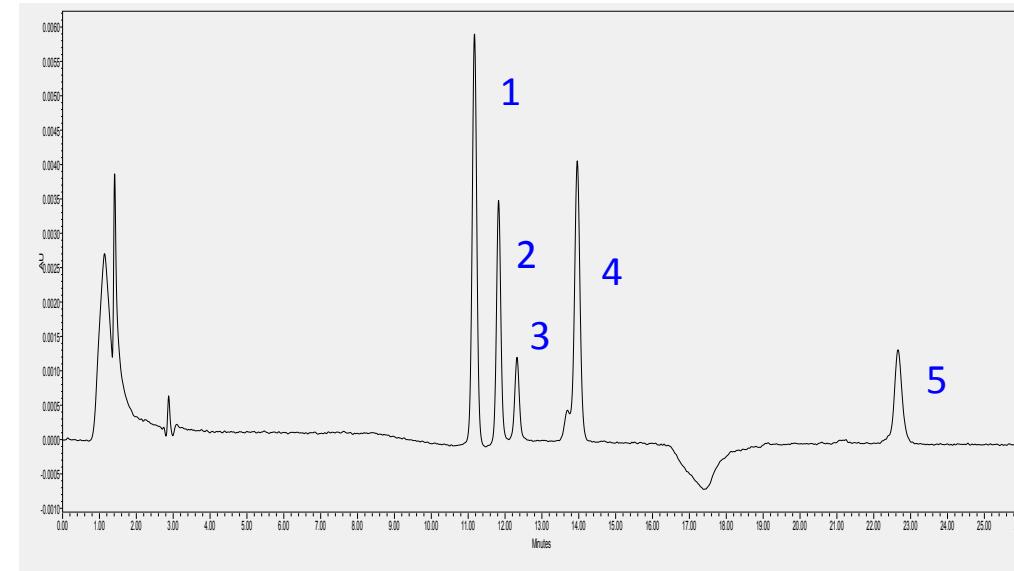
流动相： A : 0.1%乙酸水 B : 甲醇

检测波长： 325nm

流速： 1.0 ml/min

色谱柱型号： Zorbax SB-AqC18 4.6*250mm

序号	化合物
1	视黄醇
2	视黄酸
3	全反式视黄醛
4	乙酸视黄脂
5	视黄醇棕榈酸酯



流动相梯度：

Time(min)	0	5	8	13	14	24	24.5	26
A(%)	20	20	10	10	0	0	20	20
B(%)	80	80	90	90	100	100	80	80



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三、LC-QTOF的原理及应用

LC-QTOF的原理——常用术语

分辨率

质谱仪对相近质量数的分辨能力，以 $m/\Delta m$ 表示； m 为质谱峰的 m/z 值、 Δm 为该质谱峰的半峰宽。

质量准确度

质量分析器测定得到的离子质量与其质量真实值（准确质量）的接近程度。通常质量准确度用 ppm 表示。离子 m/z 需至少测定至第4位小数。

$$\text{ppm} = \text{parts per million} = m/m \times 10^6$$

例如：

真实质量 = 400.0000

测定质量 = 400.0020

误差 = 0.0020

$$\text{error} = \frac{0.002}{400} \times 10^6 = 5 \text{ ppm}$$

LC-QTOF的原理——常用术语

精确质量的重要性

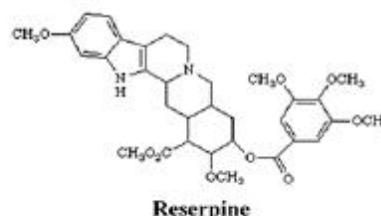
¹² C	12.0000
¹ H	1.0078
¹⁴ N	14.0031
¹⁶ O	15.9949

Reserpine ($C_{33}H_{40}N_2O_9$) has MH^+ at 609.28066 Da

Single quad gives mass to +/- 0.1 Da = 165 ppm

Number of possible elemental formulae using only C, H, O & N:

- 165 ppm 209
- 10 ppm 13
- 5 ppm 7
- 3 ppm 4
- 2 ppm 2



Accurate mass reduces number of candidates and risk of investing effort with the wrong molecule

准确质量测定增加了分子式确定的可信度

LC-QTOF的原理——结构组成

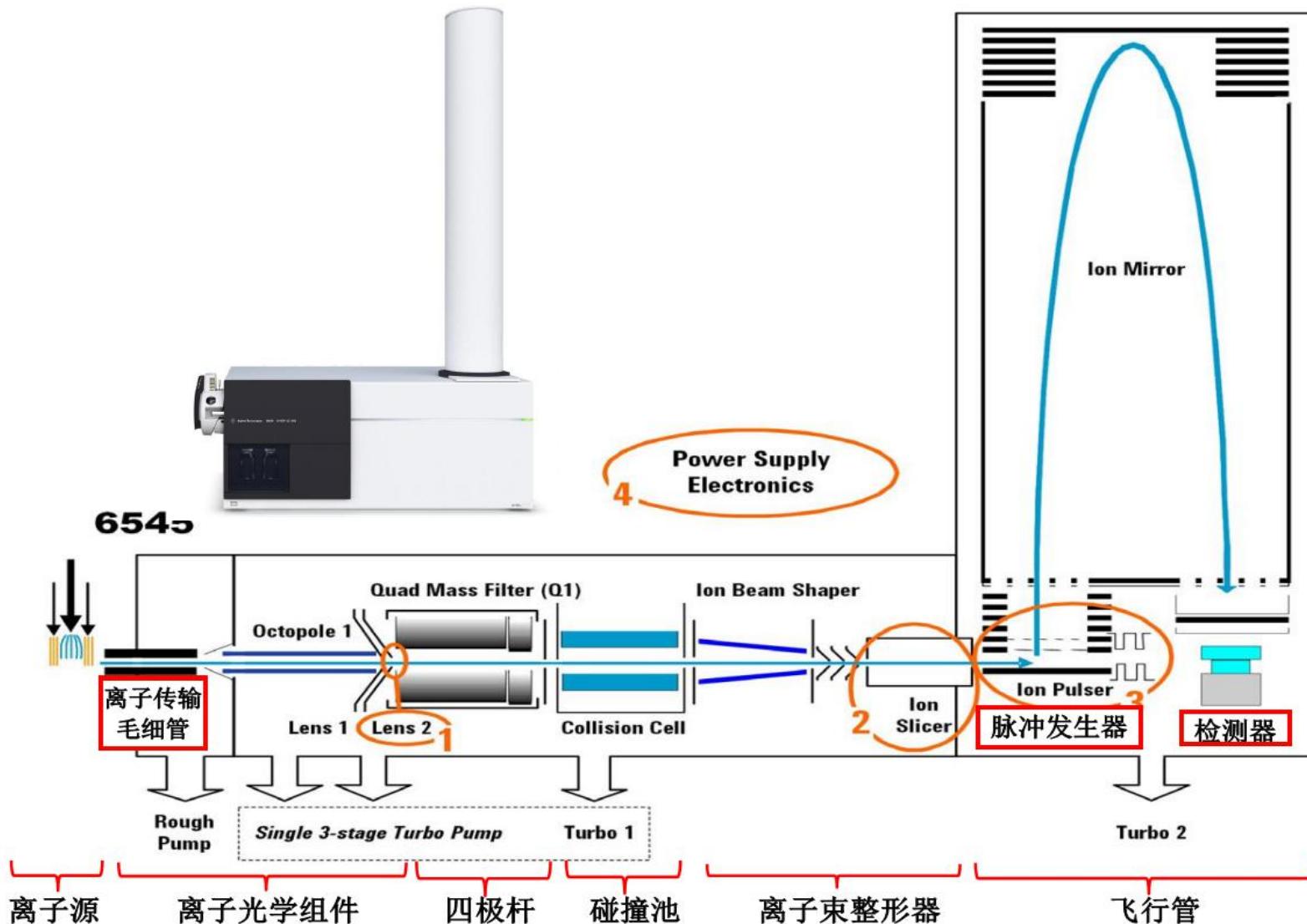


超高效液相色谱



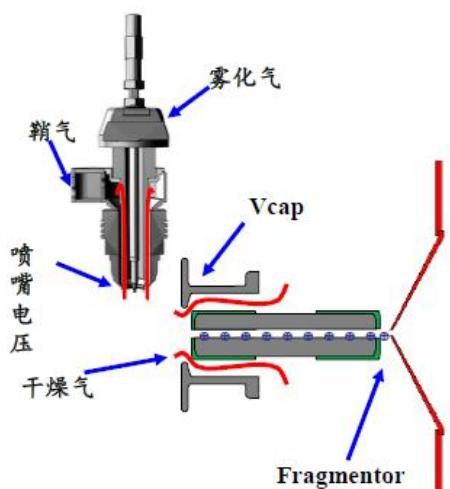
四级杆-飞行时间质谱仪

LC-QTOF的原理——结构组成

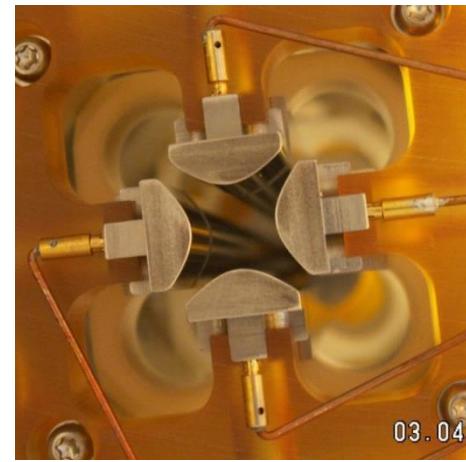


LC-QTOF的原理——核心部件

离子源



四级杆



负极杆

负极杆

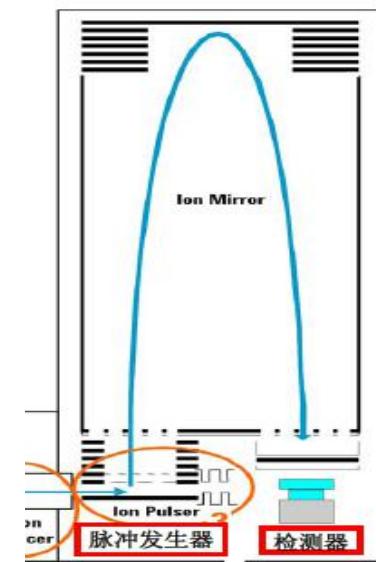
过滤高质荷比

正极杆

正极杆

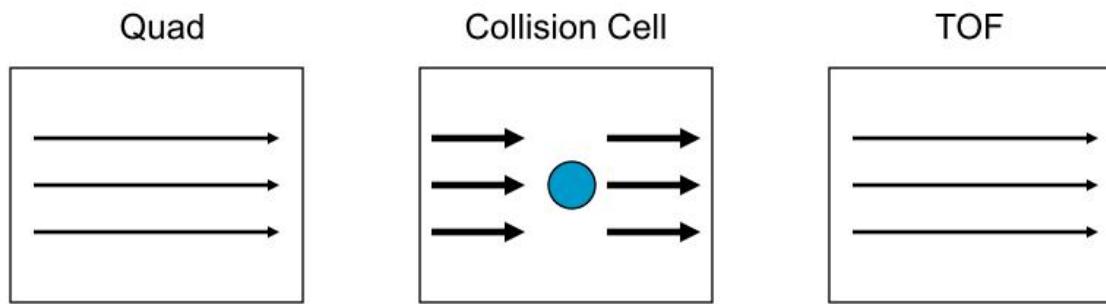
过滤低质荷比

飞行管

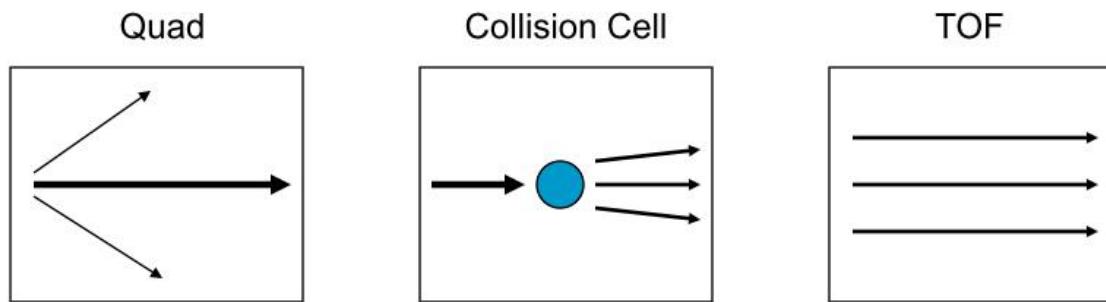


LC-QTOF的原理——工作模式

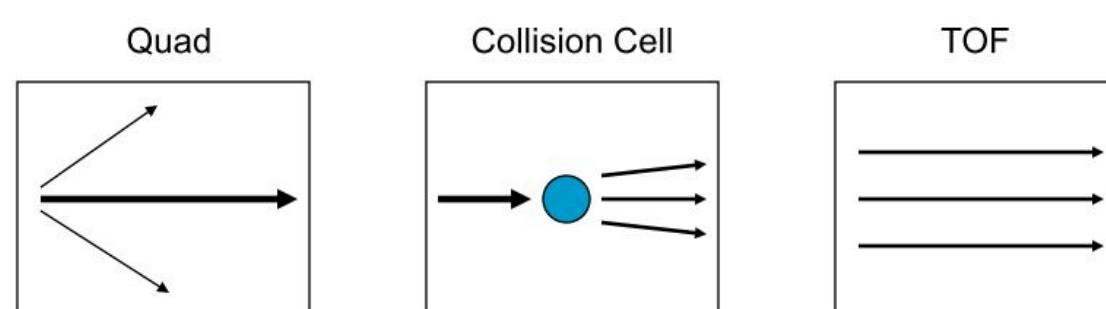
MS 模式



Auto MS/MS 模式



Targeted MS/MS 模式



LC-QTOF的应用

基础与临床研究

对与疾病相关的代谢生物标记物进行鉴定与验证，同时可有助于对生物学的基本了解

农业

确定并了解代谢途径以优化农作物开发，提高产量及杀虫剂/除草剂抗性

系统毒理学

寻找血浆和尿液中可作为药物和环境污染物接触水平替代评估指标的毒性预测性标记



食品与营养

鉴定与食品质量、真伪、风味和营养价值等主要特性相关的代谢物是否存在，并辅助保健品的开发

生物燃料与合成生物学

鉴定代谢物图谱以优化发酵过程和生物燃料生产

环境

鉴定与影响生物系统的化学物质和环境中其他刺激因子相关的代谢物

LC-QTOF的应用——化合物的筛查及定性

确定目标

了解目标物的化学性质、化学分子式、CAS号等

建数据库

利用化学分子式或标准物质谱图建立数据库

建立方法

建立合适的液相，质谱分析条件

样品筛查

检测样品，导入数据库筛查，定性或定量

LC-QTOF的应用——化合物的筛查及定性

案例一：水溶性麻痹性贝毒素的筛查

确定目标

毒素	成分	CAS注册号	分子式
麻痹性贝毒素 (水溶性)	STX	35554-08-6	$C_{10}H_{19}N_7O_4Cl_2$
	NEO-STX	64296-20-4	$C_{10}H_{17}N_7O_5$
	GTX1	60748-39-2	$C_{10}H_{17}N_7O_9S_1$
	GTX4	64296-26-0	$C_{10}H_{17}N_7O_9S_1$
	GTX2	60508-89-6	$C_{10}H_{17}N_7O_8S_1$
	GTX4	60537-65-7	$C_{10}H_{17}N_7O_8S_1$
	GTX5	64296-25-9	$C_{10}H_{17}N_7O_7S_1$

建立PCDL数据库

The screenshot shows the MassHunter PCDL Manager interface with the following details:

- Search Criteria:** The search criteria section includes fields for "Must also contain" and "Must not contain". It also includes checkboxes for "Include neutrals", "Include anions", and "Include cations". Tolerance settings for Mass (10.0 ppm), RT (0.1 min), and RI (10.00) are shown.
- Search Options:** Options for "Search only visible columns" and "Search all columns" are available, along with checkboxes for "With spectra" and "With CCS".
- Results Table:** The "Compound Results: 14 hits" table lists 14 compounds with their names, formulas, masses, retention times, and retention indices. The first three rows correspond to the target compounds from the table above:

Name	Formula	Mass	Retention Time	Retention Index	Cation	Anion	CAS	ChemSpider	PubChem
New Compound	$C_{10}H_{17}N_7O_4$	299.1342			<input type="checkbox"/>	<input type="checkbox"/>	35554-08-6		
New Compound	$C_{10}H_{17}N_7O_5$	315.12912			<input type="checkbox"/>	<input type="checkbox"/>	64296-20-4		
New Compound	$C_{10}H_{17}N_7O_9S_1$	411.08085			<input type="checkbox"/>	<input type="checkbox"/>	60748-39-2		
New Compound	$C_{10}H_{17}N_7O_9S_1$	411.08085			<input type="checkbox"/>	<input type="checkbox"/>	64296-26-0		
New Compound	$C_{10}H_{17}N_7O_8S_1$	395.08593			<input type="checkbox"/>	<input type="checkbox"/>	60508-89-6		
New Compound	$C_{10}H_{17}N_7O_8S_1$	395.08593			<input type="checkbox"/>	<input type="checkbox"/>	60537-65-7		
New Compound	$C_{10}H_{17}N_7O_7S_1$	379.09102			<input type="checkbox"/>	<input type="checkbox"/>	64296-25-9		
New Compound	$C_{55}H_{82}O_{21}S_2$	1142.4790			<input type="checkbox"/>	<input type="checkbox"/>			
New Compound	$C_{56}H_{84}O_{21}S_2$	1156.49465			<input type="checkbox"/>	<input type="checkbox"/>			
New Compound	$C_{45}H_{86}O_{10}$	786.6221			<input type="checkbox"/>	<input type="checkbox"/>			
New Compound	$C_{51}H_{96}O_{15}$	948.67492			<input type="checkbox"/>	<input type="checkbox"/>			
New Compound	$C_{45}H_{70}O_{13}$	818.49164			<input type="checkbox"/>	<input type="checkbox"/>			
New Compound	$C_{50}H_{70}O_{14}$	894.47656			<input type="checkbox"/>	<input type="checkbox"/>			
New Compound	$C_{50}H_{72}O_{14}$	896.49221			<input type="checkbox"/>	<input type="checkbox"/>			

LC-QTOF的应用——化合物的筛查及定性

液相检测条件：

流动相： A : 0.1% Fomic Acid Water

B : 0.1% Fomic Acid ACN

流速： 0.3 ml/min

色谱柱型号： Waters BEH amide, 2.1X100 mm; 1.7 um

6545 QTOF质谱条件：

Ion Source: AJS

Polarity : Positive

Gas temp : 320 °C

Drying Gas: 8 L/min

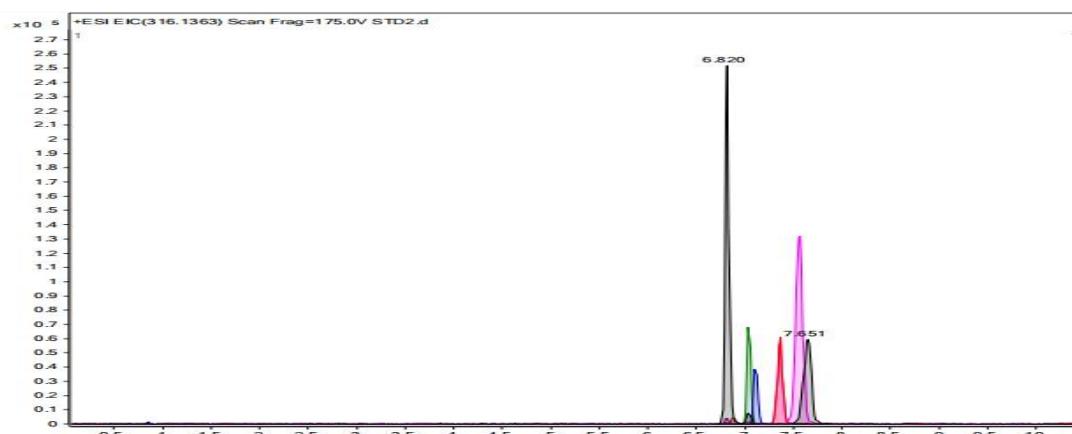
Nebulizer: 35 psig

VCap: 3500 V

Sheath gas temp : 350 °C

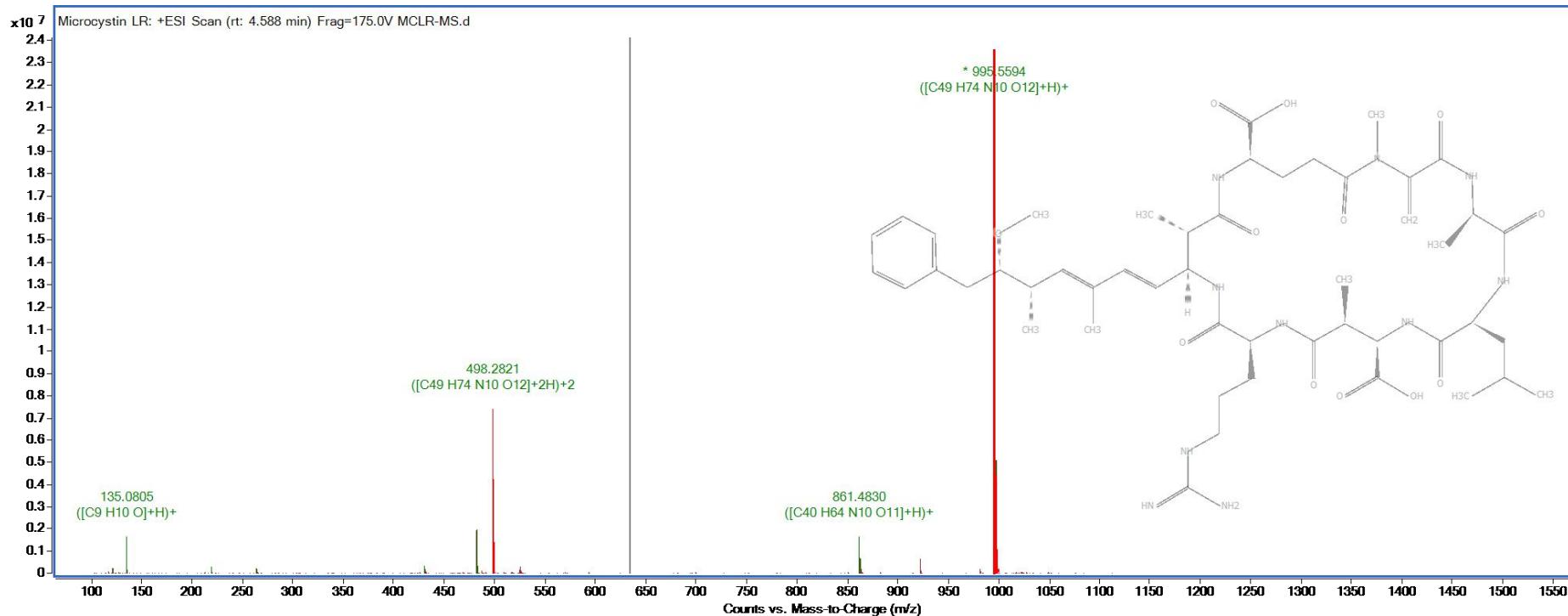
Sheath gas flow: 11 L/min

流动相梯度 : Time(min)	0	2	6	8	8.5	10.5
A(%)	10	10	43	43	10	10
B(%)	90	90	57	57	90	90



LC-QTOF的应用——化合物的筛查及定性

案例二：合成MC-LR的定性



LC-QTOF的应用——靶向代谢组学的研究

脂质组学 - 了解疾病发病机理，寻找生物标识物，开发治疗途径

> 医科院药物所 6410/6490 QQQ & 6550 QTOF

> 靶向脂质组学 - 迟发型超敏反应生物标识物/儿童皮炎

43种主要鞘酯靶向检测

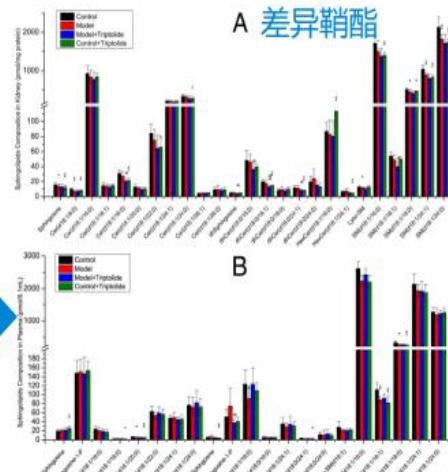
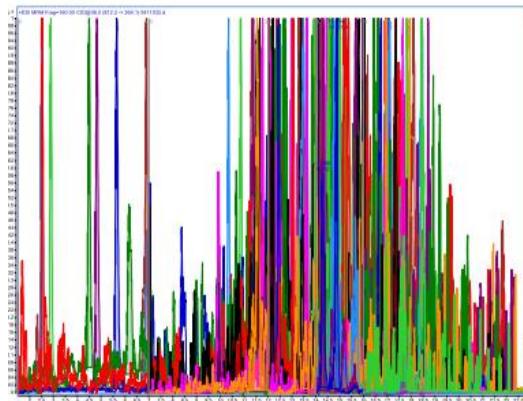


Figure 4. Sphingolipid composition of mouse kidney or plasma measured by triple quadrupole MS/MS. Four groups including control, model, model+triprolide and control+triprolide, each of which contains 8 samples. Sphingolipids were isolated from kidney homogenate (0.5 g) or plasma (0.1 ml), then extracted and analyzed by LC-QTOF-MS/MS. The relative mass was overlaid by the fold changes in sphingolipid metabolites between the control group and other groups by asterisks indicating the statistical significance on the right side (* = P<0.05, ** = P<0.01; n = 8). The content of each sphingolipid metabolite was determined as the mean of eight independent parallel samples.

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PLOS ONE

Sphingolipids as New Biomarkers for Assessment of Delayed-Type Hypersensitivity and Response to Triptolide

Feng Qu, Cai-Sheng Wu, Jin-Feng Hou, Ying Jin, Jin-Lan Zhang*

State Key Laboratory of Bioactive Substance and Function of Natural Medicines, Institute of Materia Medica, Peking Union Medical College and Chinese Academy of Medical Sciences, Beijing, China

Abstract

Background: Hypersensitivity diseases are associated with many severe human illnesses, including leprosy and tuberculosis. Emerging evidence suggests that the pathogenesis and pathological mechanisms of treating these diseases may be attributable to sphingolipid metabolism.

Methods: High performance liquid chromatography-tandem mass spectrometry was employed to target and measure 43 core sphingolipids in the plasma, kidneys, livers and spleens of BALB/c mice from four experimental groups: control, delayed-type hypersensitivity (DTH) model, DTH+triprolide, and control+triprolide. Orthogonal partial least squares discriminant analysis (OPLS-DA) was used to identify potential biomarkers associated with variance between groups. Relationships between the identified biomarkers and disease markers were evaluated by Spearman correlation.

Results: As a treatment to hypersensitivity disease, triptolide significantly inhibit the ear swelling and recover the reduction

鞘酯生物合成通路

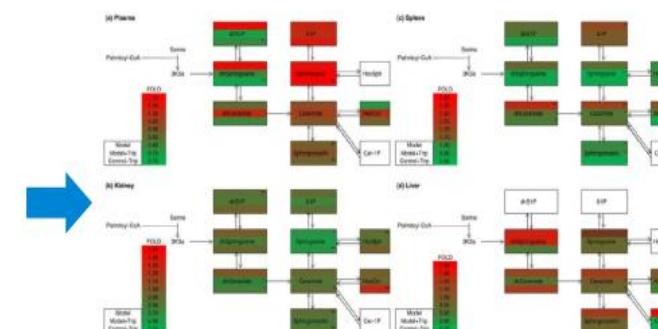


Figure 4. Visualization of sphingolipid metabolites in the plasma, kidney, spleen and liver. The figure depicts sphingolipid metabolites that participate in the early steps of sphingolipid biosynthesis. The relative mass are overlaid by the fold changes in subclasses of sphingolipid metabolites between the control group and other groups by asterisks indicating the statistical significance on the right side (* = P<0.05, ** = P<0.01; n = 8). The content of each sphingolipid metabolite was determined as the mean of eight independent parallel samples.

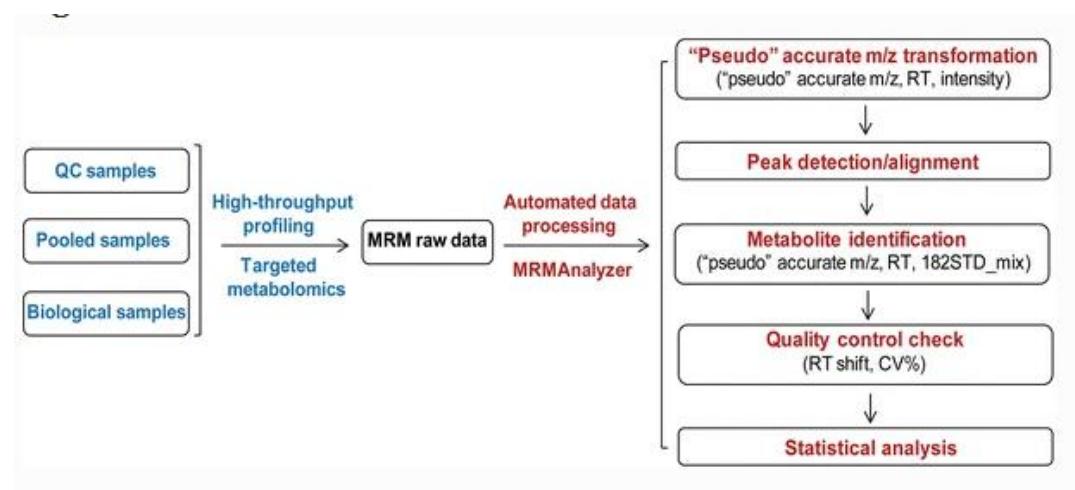
LC-QTOF的应用——靶向代谢组学的研究

- 上海有机所生物与化学交叉研究中心6460QTOF
- 代谢组学/脂质组学检测平台

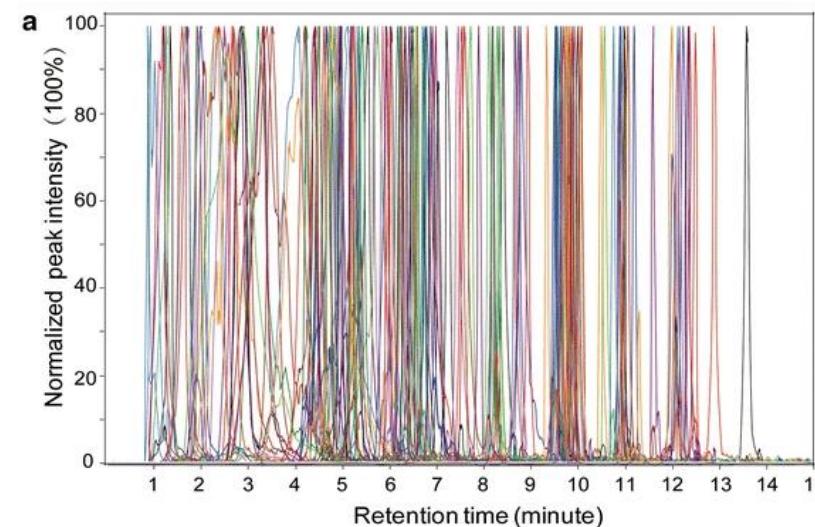
An integrated targeted metabolomic platform for high-throughput metabolite profiling and automated data processing

[Yuping Cai, Kai Weng, Yuan Guo, Jie Peng & Zheng-Jiang Zhu](#)✉

[Metabolomics](#) 11, 1575–1586(2015) | [Cite this article](#)

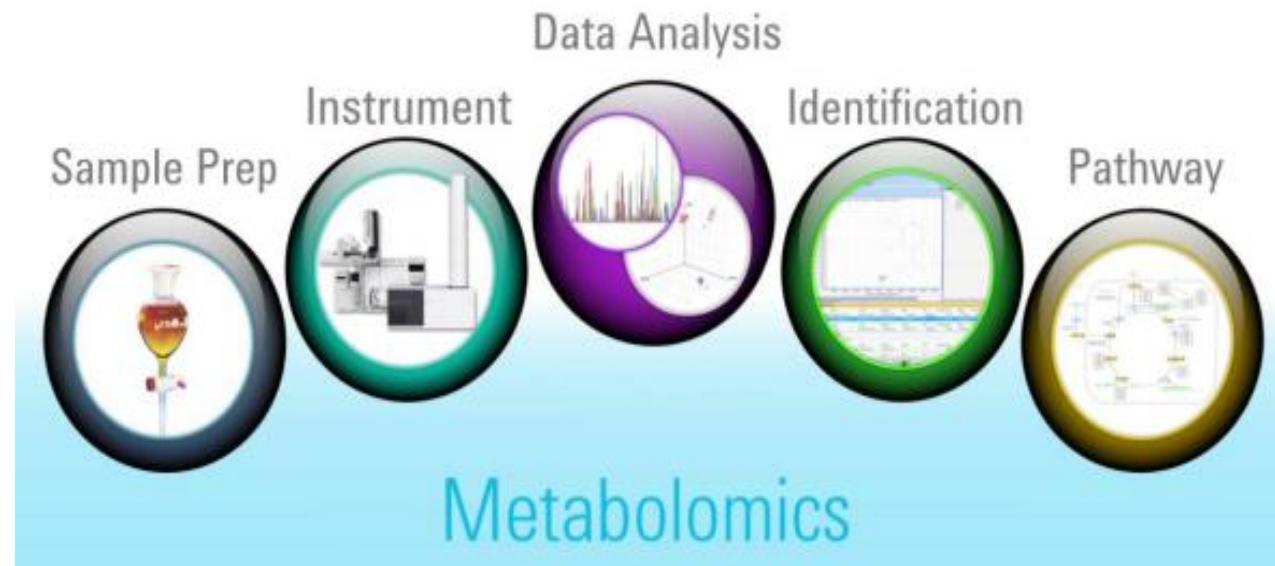


182种主要代谢产物同时检测



LC-QTOF的应用——非靶向代谢组学的研究

非靶向代谢组学分析流程



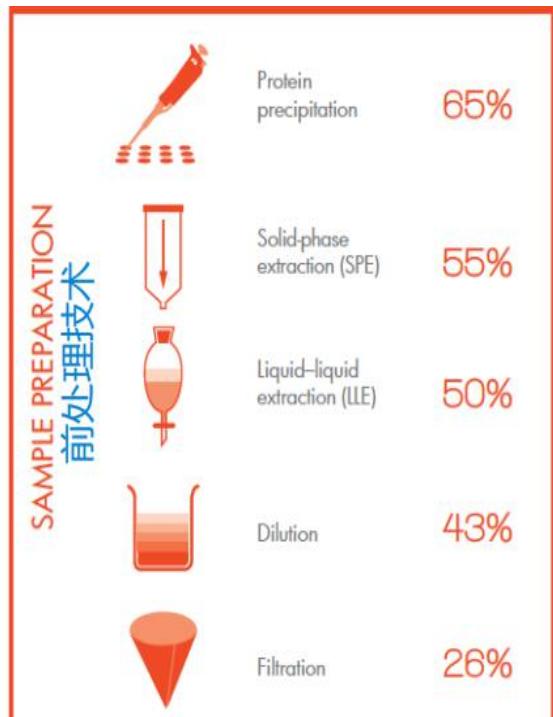
LC-QTOF的应用——非靶向代谢组学的研究

实验设计

1. 明确要解决的问题；
2. 选择合适的生物模型系统（例如人类、大鼠等）；
3. 选择正确的生物体液、组织、细胞；
4. 足够的生物性重复样本，以消除内在变量的影响；
5. 选择合适的分析仪器，如LC-MS/GC-MS/LC-QTOF MS;
6. 充足合适的实验耗材，如色谱柱、样品管、试剂。

LC-QTOF的应用——非靶向代谢组学的研究

样品制备



1. 血浆/血清

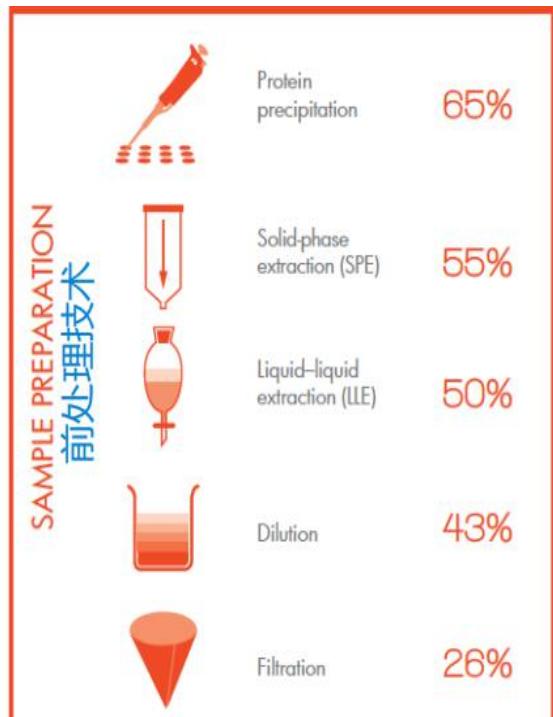
取 100 μ L 血清/血浆样品置于 1.5mL 离心管中，加入 400 μ L 甲醇/乙腈混合溶剂(1:1, v/v)，涡旋 30 秒，4°C 水浴中超声 10 分钟后，在-20°C 冰箱中静置 1 小时；取出离心管在 4°C 下 12000rpm 离心 15 分钟，取上清液，真空离心旋干；旋干后样品用 100 μ L 水/乙腈混合溶剂(1:1, v/v)复溶，4°C 水浴中超声 10 分钟，4°C 下 12000rpm 离心 15 分钟，取上清液-80°C 保存至检测。

2. 动物组织

取液氮淬灭后组织样品(大于 10mg)，加入 1mL 甲醇/乙腈/水混合溶剂(2: 2: 1, v/v)匀浆，在液氮、37°C 水浴中反复冻融 3 次；将样品在-20°C 下静置 1 小时后，在 4°C 下 13000rpm 离心 15 分钟，取上清液，真空离心旋干；旋干后样品用 100 μ L 水/乙腈混合溶剂(1:1, v/v)复溶，4°C 水浴中超声 10 分钟，4°C 下 13000rpm 离心 15 分钟，取上清液-80°C 保存至检测。

LC-QTOF的应用——非靶向代谢组学的研究

样品制备



3.细胞/细菌

取一盒细胞(约 $10^6\text{-}10^7$ 个),倒掉培养基,用10mL冰磷酸盐缓冲液(PBS)清洗两遍,液氮淬灭;加入1mL甲醇/乙腈/水混合溶剂(2:2:1, v/v),用细胞刮刀刮下细胞,置于1.5mL离心管中;涡旋30秒后,在液氮、37℃水浴中反复冻融3次;将样品在-20℃下静置1小时后,在4℃下13000rpm离心15分钟,取上清液,真空离心旋干;旋干后样品用100μL水/乙腈混合溶剂(1:1, v/v)复溶,4℃水浴中超声10分钟,4℃下13000rpm离心15分钟,取上清液-80℃保存。

4.植物组织

植物组织经液氮速冻后均匀研碎成粉末,称取100mg粉末样品置于1.5mL离心管中,加入预先冷冻的含0.125%甲酸的甲醇溶液300μL,涡旋10秒,在20℃水浴下超声15分钟,13000rpm离心15分钟,取上清液-80℃保存至检测。

LC-QTOF的应用——非靶向代谢组学的研究

仪器分析

代谢组学分析常用仪器特点:

➤ GC-MS

- 易挥发，低极性，热稳定的小分子化合物
- 需衍生化

➤ LC-MS

- 具有一定极性的有机化合物
- 无需衍生化

➤ CE-MS

- 高极性化合物，如核酸，蛋白等

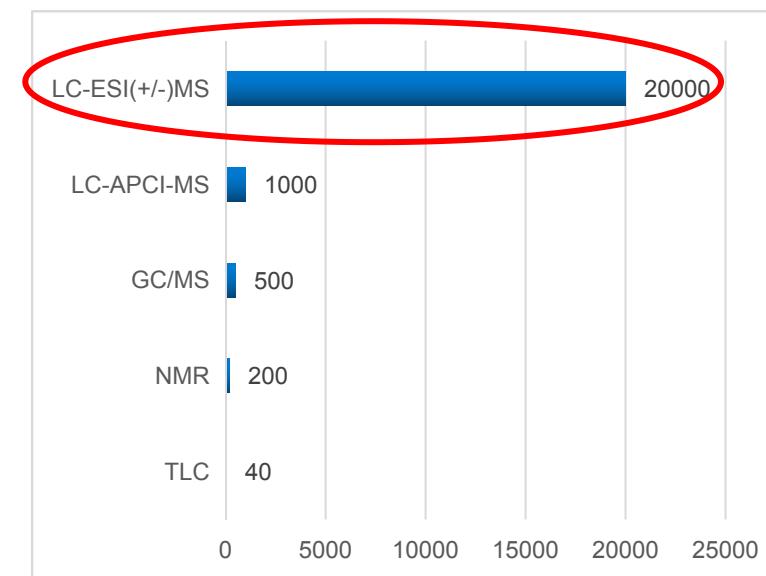
➤ ICP-MS

- 无机化合物

➤ NMR

- 无偏性，无损检测
- 无需繁琐前处理，便于活体、原位的动态检测

代谢组学分析技术及各技术可以检测到化合物数量



LC-QTOF的应用——非靶向代谢组学的研究

数据处理

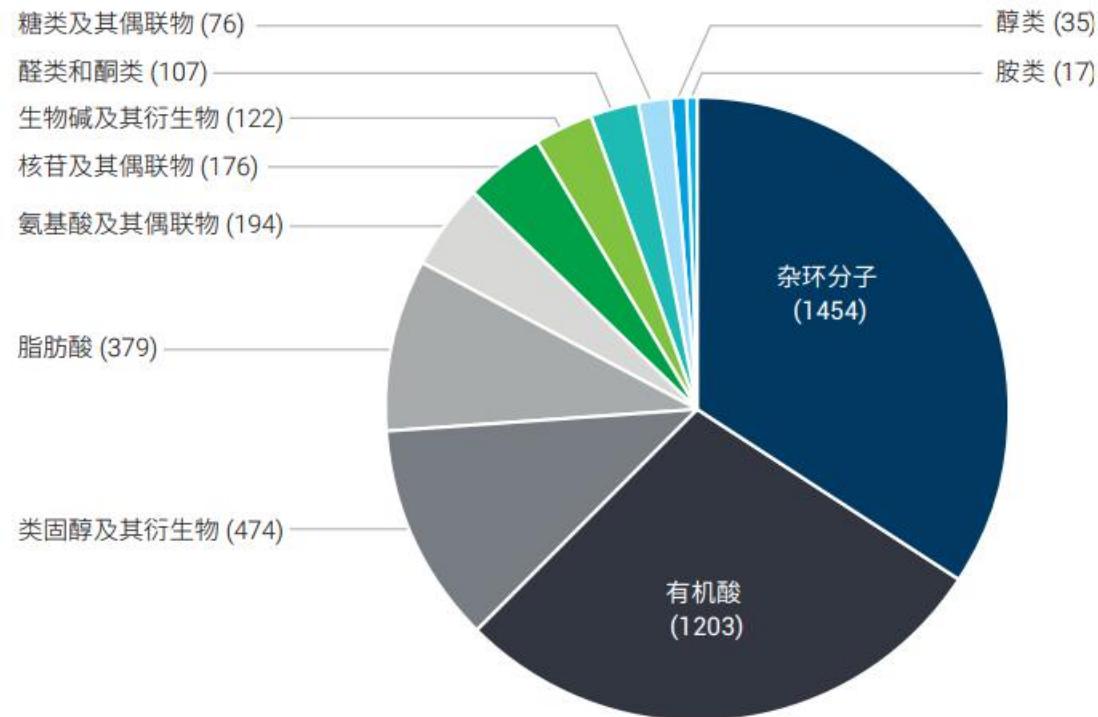
MassHunter Profinder – 多数据质谱特征提取工具



LC-QTOF的应用——非靶向代谢组学的研究

数据处理

Agilent METLIN代谢组学数据库



收录80000+生物分子的数据库，其中10000+生物分子有使用标品采集得到的谱图。

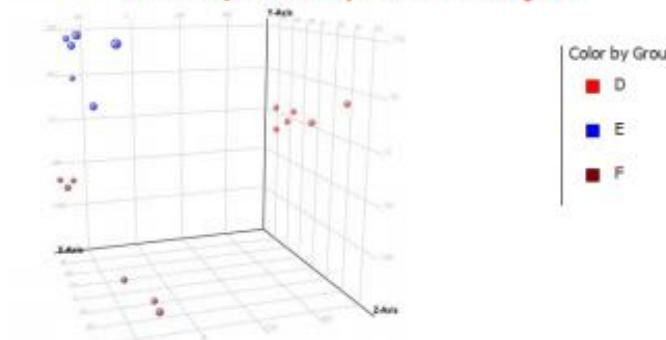
LC-QTOF的应用——非靶向代谢组学的研究

数据处理

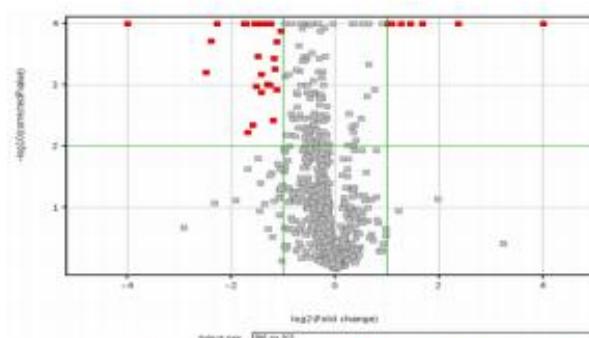
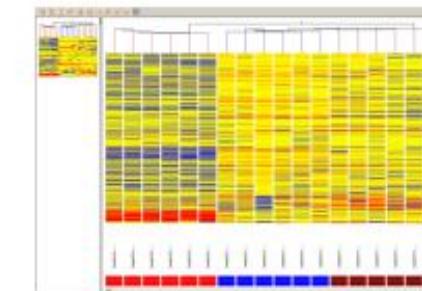
MPP: 丰富的统计学算法

(ANOVA, Student t-test, Clustering, PCA, PLS-DA, Correlation, Class prediction tools, etc)

Principle Component Analysis

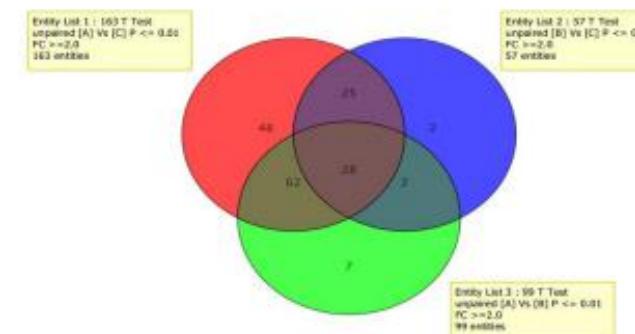


Unsupervised Hierarchical Clustering



1-way Anova, FC > 2, P < 0.05

Venn Diagram of Fold Changes > 2



Entity List 1 : 163 T Test unpaired [A] Vs [C] P <= 0.05
FC >=2.0
163 entities

Entity List 2 : 57 T Test unpaired [B] Vs [C] P <= 0.05
FC >=2.0
57 entities

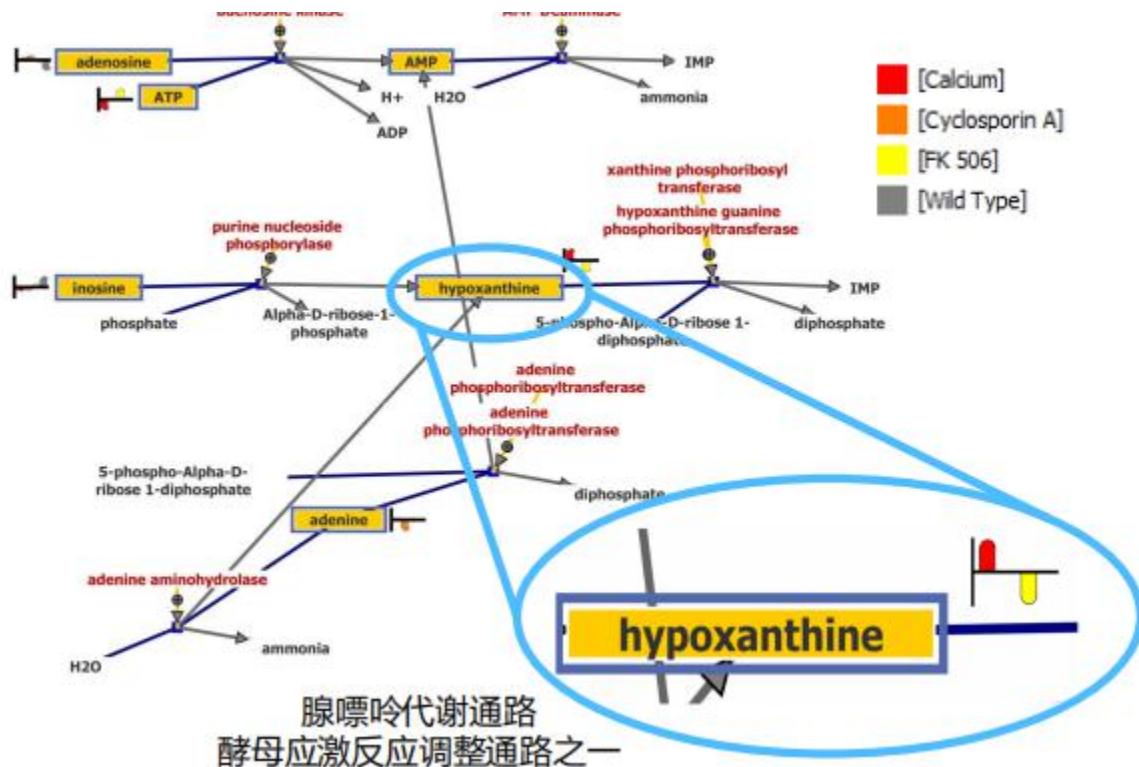
Entity List 3 : 99 T Test unpaired [A] Vs [B] P <= 0.05
FC >=2.0
99 entities

LC-QTOF的应用——非靶向代谢组学的研究

数据处理

通路分析

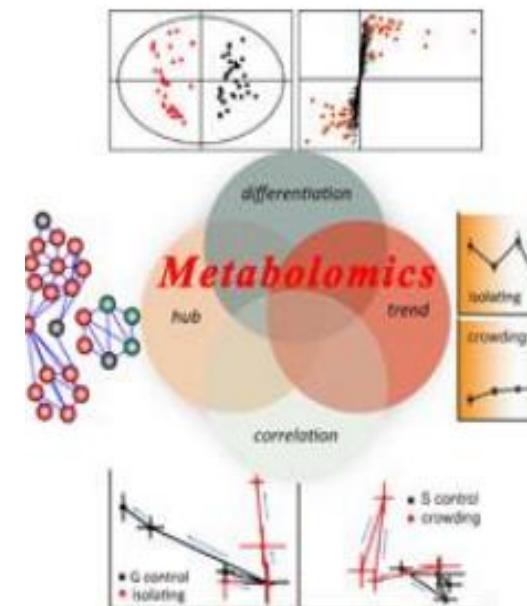
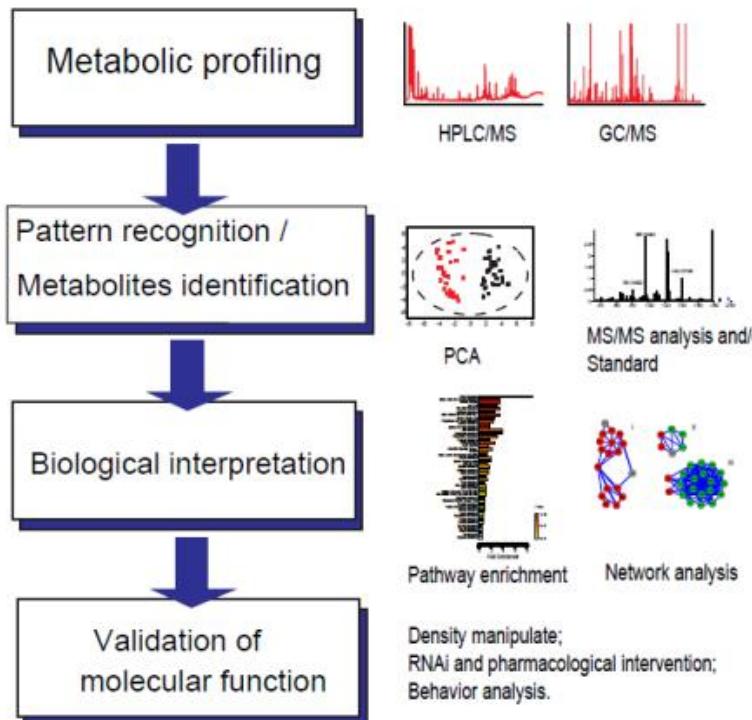
- 在本地实现生物通路的检索，浏览和过滤。
- 可视化通路结果
- 支持单组学和多组学通路分析
- 通路来源于主流的通路数据库
 - WikiPathways
 - BioCyc
 - KEGG
 - 用户自定义通路
- 支持通路中化合物导出



LC-QTOF的应用——非靶向代谢组学的研究

基础研究 - 了解生命过程

- > 中科院动物所与大连化物所 6510 QTOF & MPP
- > 首次发现了肉碱类代谢物在飞蝗两型转变过程中的关键调控作用



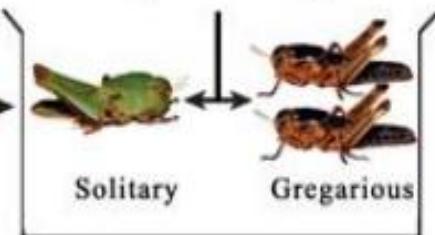
PNAS

Metabolomic analysis reveals that carnitines are key regulatory metabolites in phase transition of the locusts

Rui Wu^{a,1}, Zeming Wu^{b,1}, Xianhui Wang^{b,1}, Pengcheng Yang^a, Dan Yu^a, Chunxia Zhao^b, Guowang Xu^{b,2}, and Le Kang^{a,2}
^aState Key Laboratory of Integrated Management of Pest Insects and Rodents, Institute of Zoology, Chinese Academy of Sciences, Beijing 100101, People's Republic of China; and ^bChinese Academy of Sciences Key Laboratory of Separation Science for Analytical Chemistry, National Chronotographic Research and Analysis Centre, Dalian Institute of Chemical Physics, Chinese Academy of Sciences, Dalian 110023, People's Republic of China

Edited by David L. Denlinger, Ohio State University, Columbus, OH, and approved January 13, 2012; revised for review November 22, 2011.
Phenotypic plasticity occurs prevalently and plays a vital role in adaptive evolution. However, the underlying molecular mechanisms responsible for the expression of alternate phenotypes remains unknown. Here, a density-dependent phase polyphenism of *Locusta migratoria* was used as the study model to identify key signaling molecules regulating the expression of phenotypic plasticity. Metabolomic analysis, using high-performance liquid chromatography and gas chromatography-mass spectrometry, showed that solitary and gregarious locusts have distinct metabolic profiles in hemolymph. A total of 319 metabolites, many of which are involved in lipid metabolism, differed significantly in energy metabolism, which do not all share the same underlying mechanisms (γ). The transition between solitary and gregarious phases is a reversible process and continues even across generations. Behavioral change has been regarded as a key step for the establishment of a positive-feedback loop that can drive an initially solitary phase to the gregarious phase, which is triggered by multiple sensory inputs from other individuals (γ). The neuromodulation of CNS circuits and the changes of olfactory sensitivity have been recently proposed to be involved in mediating the initiation of behavioral phase change (δ - β).

Carnitines are key regulators of phase change!



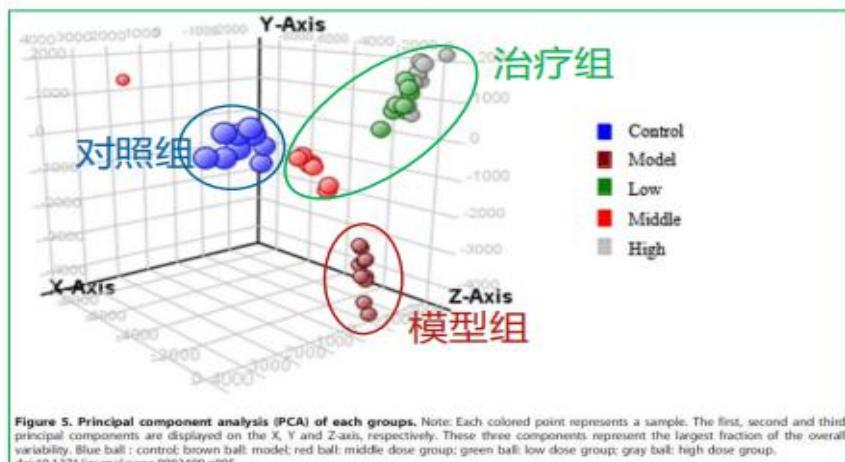
LC-QTOF的应用——非靶向代谢组学的研究

PLOS ONE

临床研究/药物研发

> 6220 TOF-MS & MPP

> 胃溃疡的生物标识物及延胡索生物碱的治疗机理研究



胃溃疡的生物标识物

Table 1. The identified potential biomarkers and metabolic pathway between groups.

RT	m/z	Molecular formula	metabolites	Metabolic pathway
1	1.018	C ₆ H ₁₂ O ₆	D-glucose	glucuronidation
2	1.021	C ₆ H ₁₄ N ₂ O ₂	L-Lysine	Biotin metabolism
3	1.063	C ₅ H ₄ N ₂ O ₃	Uric acid	Folic acid network
4	1.128	C ₃ H ₆ O ₃	Pyruvic acid	Glycolysis and gluconeogenesis
5	1.441	C ₁₁ H ₁₁ N ₂ O ₂	D-Tryptophan	Folic acid network
6	3.588	C ₂₈ H ₄₁ NO ₆	Glycocholate	Fatty acid biosynthesis
7	4.964	C ₂₁ H ₃₀ O ₄	corticosterone	Biosynthesis of aldosterone and cortisol
8	5.188	C ₁₈ H ₄₀ NO ₃ P	sphingosine-1-phosphate	Sphingolipid metabolism
9	6.132	C ₁₆ H ₃₀ O ₄	hexadecanedioic acid	Fatty acid biosynthesis
10	9.363	C ₁₈ H ₃₆ O ₂	stearic acid	Fatty acid biosynthesis

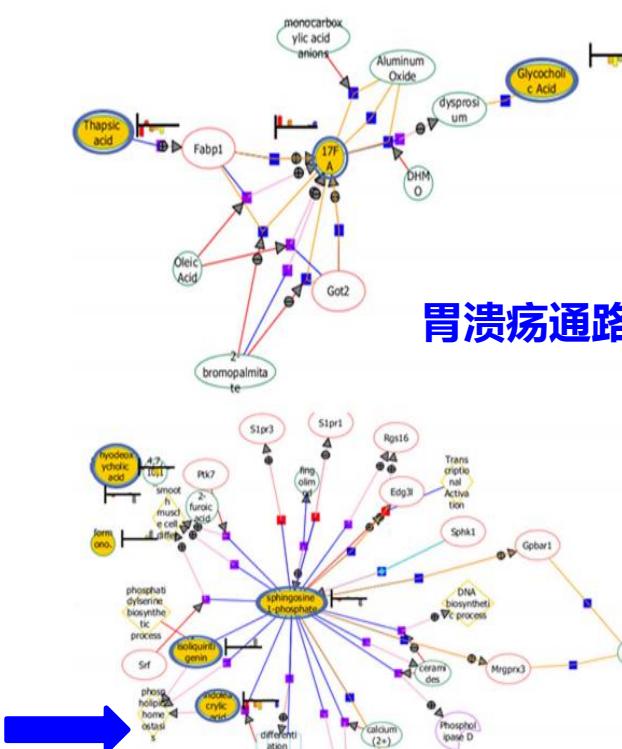


Figure 8. The network of gastric ulcer. Note: Yellow points in the fig represent biomarkers that have significant difference between the metabolites. The multi-point and multi-pathway involve in combined effect to the formation and healing of gastric ulcer.

OPEN ACCESS Freely available online

Metabolomics Coupled with Multivariate Data and Pathway Analysis on Potential Biomarkers in Gastric Ulcer and Intervention Effects of *Corydalis yanhusuo* Alkaloid

Li Tianjiao^{1,*}, Wang Shuai^{1,3}, Meng Xiansheng^{1,3,✉}, Bao Yongrui¹, Guan Shanshan¹, Liu Bo¹, Chen Lu¹, Wang Lei^{2,1}, Ran Xiaorong^{2,3*}

¹ Liaoning University of Traditional Chinese Medicine, Dalian, P.R. China, ² Agilent Technologies Co., Ltd, Beijing, P.R. China, ³ Liaoning University of Traditional Chinese Medicine-Agilent Technologies Modern TCM and Multi-omics Research Collaboration Lab, Dalian, P.R. China

Abstract

Metabolomics, the systematic analysis of potential metabolites in a biological specimen, has been increasingly applied to discovering biomarkers, identifying perturbed pathways, measuring therapeutic targets, and discovering new drugs. By analyzing and verifying the significant difference in metabolic profiles and changes of metabolite biomarkers, metabolomics enables us to better understand substance metabolic pathways which can clarify the mechanism of Traditional Chinese Medicine(TCM). *Corydalis yanhusuo* alkaloid (CYA) is a major component of Qizhiweiutong (QZWT) prescription which has been used for treating gastric ulcers for centuries and its mechanism remains unclear completely. Metabolite profiling was performed by high-performance liquid chromatography combined with time-of-flight mass spectrometry (HPLC/ESI-TOF-MS).

转录组实验验证

胃溃疡通路构建

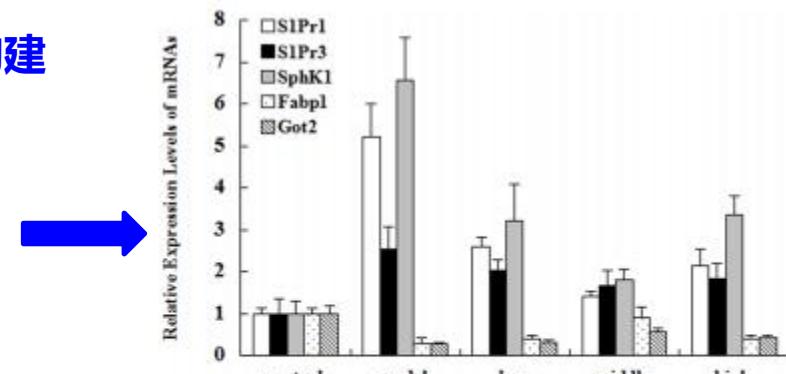


Figure 9. The expression level of mRNAs. Note: Abscissa represents the mRNAs of sphingolipid metabolism (including S1Pr1, S1Pr3 and SphK1) and fatty acid metabolism (including fabp1 and got2). The method of relative quantitative analysis was used to compare the gene expression in each group. The ordinate represents the relative expression levels of mRNAs in the basis of control group. Quantitative PCR results represent mean ± SEM of three independent experiments.



中国科学院水生生物研究所

INSTITUTE OF HYDROBIOLOGY, CHINESE ACADEMY OF SCIENCES



中科院水生所

四、平台成员及服务内容

平台成员及服务内容



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服务一：化合物的测试

有机化合物

无机化合物

元素

服务二：实验操作培训

GCMS

HPLC-MS

ICPMS

ICPOES

IRMS

服务三：实验方案规划

实验前：采样/样品准备

实验中：仪器的选择

实验后：数据处理



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INSTITUTE OF HYDROBIOLOGY, CHINESE ACADEMY OF SCIENCES

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谢 谢

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