

BIOCRATES

LIFE SCIENCES

The Deep Phenotyping Company

Worldwide First, Quantitative Biocrates[®] Bile Acids Kit Standardized LC-MS/MS Based Analysis



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Mass Spectrometry Based Assay for the Quantitation of
Endogenous Bile Acids in Blood

For research use only (RUO)

Robust Bile Acids Phenotyping in Blood

The **Biocrates® Bile Acids Kit*** provides ready-to-use components for an LC-MS/MS-based assay and enables the quantitative measurement of a number of bile acids in human plasma/serum (16 human specific analytes) and mouse plasma samples (19 mouse specific analytes).

- **Ready to use LC-MS/MS Kit for all major mass spectrometric platforms with standard ESI ion source**
- **Robust, fast, simple and reliable sample preparation**
- **Primary/Secondary bile acids**
- **Conjugated/Unconjugated bile acids**

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The **Bile Acids Kit** is characterized by an easy to perform, quick and highly effective sample extraction on a 96-well patented filter plate. The robust sample preparation produces clean extracts which guarantee longer performance of the analytical column and the high accuracy and precision of the analysis. The analytical column (to be ordered separately), therefore, can be used for a number of Kits. The column is applicable for both HPLC and UHPLC conditions.

To ensure the accurate and precise determination of bile acids, the Kit provides seven calibration standards, a mixture of 10 isotope labeled internal standards and three levels of quality control samples.

No	Analyte	Name	Internal Standard	Human plasma/serum	Mouse plasma	Calibration range (LLOQ – ULOQ in µM)
1	CA	Cholic acid	d5-CA	✓	✓	0.03 – 75
2	CDCA	Chenodeoxycholic acid	d5-CDCA	✓	✓	0.02 – 30
3	DCA	Deoxycholic acid	d5-CDCA	✓	✓	0.02 – 10
4	GCA	Glycocholic acid	d5-GCA	✓	✓	0.03 – 75
5	GCDCA	Glycochenodeoxycholic acid	d4-GLCA	✓		0.02 – 20
6	GDCA	Glycodeoxycholic acid	d4-GLCA	✓	✓	0.01 – 10
7	GLCA	Glycolithocholic acid	d4-GLCA	✓	✓	0.01 – 5
8	GUDCA	Glycoursodeoxycholic acid	d4-GUDCA	✓	✓	0.01 – 10
9	HDCA	Hyodeoxycholic acid	d4-HDCA(b)		✓	0.01 – 5
10	LCA	Lithocholic acid	d4-LCA	✓	✓	0.01 – 5
11	MCA(a)	Alpha-Muricholic acid	d5-CA		✓	0.01 – 5
12	MCA(b)	Beta-Muricholic acid	d5-CA		✓	0.01 – 10
13	MCA(o)	Omega-Muricholic acid	d5-CA		✓	0.01 – 5
14	TCA	Taurocholic acid	d5-TCA	✓	✓	0.02 – 50
15	TCDCA	Taurochenodeoxycholic acid	d5-TCDCA	✓	✓	0.01 – 20
16	TDCA	Taurodeoxycholic acid	d5-TCDCA	✓	✓	0.01 – 10
17	TLCA	Taurolithocholic acid	d4-GLCA	✓	✓	0.01 – 5
18	TMCA (a+b)	Tauromuricholic acid (sum of alpha and beta)	d5-TUDCA	✓	✓**	0.01 – 10
19	TUDCA	Tauroursodeoxycholic acid	d5-TUDCA	✓	✓*	0.01 – 15
20	UDCA	Ursodeoxycholic acid	d4-HDCA(b)	✓	✓	0.02 – 30

* Partial coelution with THDCA under UHPLC conditions, quantifiable only under HPLC conditions

** semi-quantitative for Waters Xevo™ TQ MS

✓ Generally present at very low concentrations (close to or < LLOQ) in healthy samples

✓ Generally present concentrations well above LLOQ in healthy samples

Kit Parameters

Total Runtime (for 96 tests):

- UHPLC: 14 hours (2 hours sample prep + 10 hours LC-MS/MS + 2 hours data processing)
- HPLC: 24 hours (2 hours sample prep + 20 hours LC-MS/MS + 2 hours data processing)

Sample matrix:

- Human plasma, human serum and mouse plasma (other matrix in progress)

LC-MS/MS Platforms:

- AB SCIEX 4000 QTRAP®, QTRAP® 5500
- Waters Xevo™ TQ MS
- Thermo TSQ Vantage™
- Other platforms in progress

Ionization:

- ESI negative

Sample preparation:

- Direct extraction on Biocrates 96-well Kit filter plate, ca. 2 hours
- 96 tests / Kit

Sample volume:

- 10 µL (20 µL for Waters Xevo™ TQ MS)

Analysis time:

- HPLC: 11 min / sample
- UHPLC : 5 min / sample

Quantitation:

- 7 external calibration standards
- 10 isotope labeled internal standards

Quality controls:

- 3 levels: low, medium, high
- Human plasma based

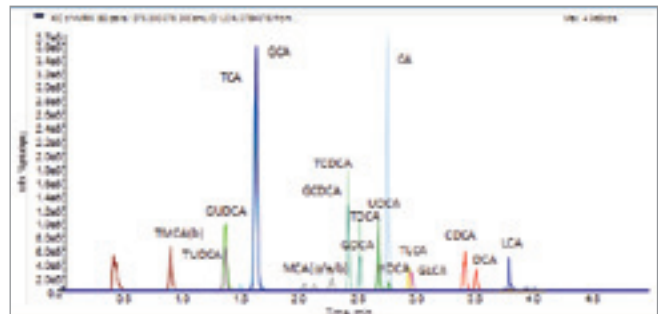


Figure 1: Chromatogram of a bile acids calibration standard measured on a UHPLC Nexera X2 (Shimadzu) coupled with a QTRAP® 5500 mass spectrometer (AB SCIEX).

Bile Acids Phenotyping

Human plasma/serum sample

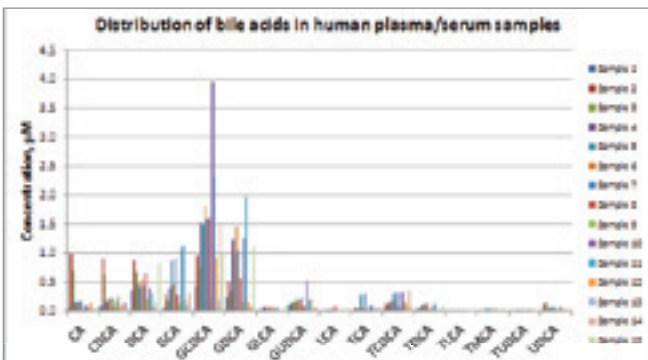


Figure 2: Distribution of bile acids in human plasma/serum samples.

Mouse plasma: male vs. female

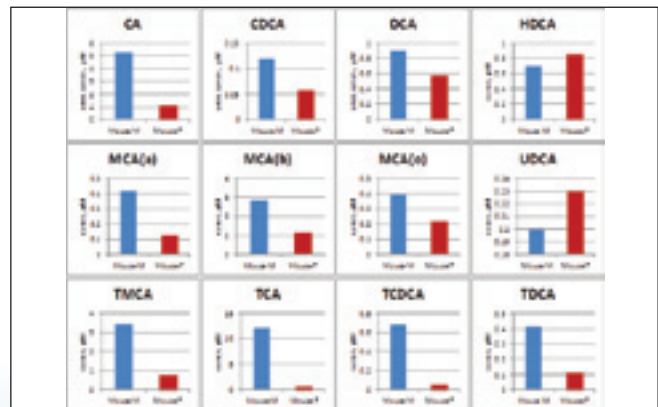


Figure 4: Differentiation between mouse male and mouse female using bile acid profile in plasma samples.

Mouse plasma vs. Human plasma/serum

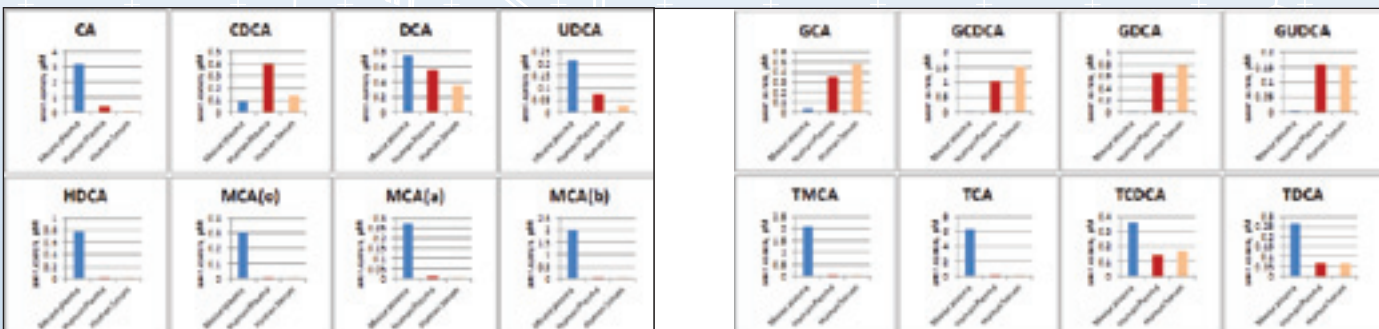


Figure 3: Differences in bile acid content between mouse plasma and human plasma/serum samples.



Kit Content

Kit Item	Description
7 level calibration standards: mixtures of 20 bile acids	7 vials Cover also abnormal range in samples Lyophilized
Isotope labeled internal standards (ISTD): mixture of 10 compounds	1 vial Dried
Testmix: mixture of 20 bile acids	2 vials Dried
3 level quality controls	3 vials Human plasma based Lyophilized
USB drive	User Manual and Analytical Specifications in pdf format
Biocrates 96-well Kit filter plate in combination with deep-well extraction receiving plate	For sample preparation
Silicone mat	For covering the deep-well receiving plate after sample preparation
Bile Acids Kit column set	Set-up set containing UHPLC analytical column, precolumn and holder (to be ordered separately)



BIOCRATES Life Sciences AG

Ordering Information

For Further Information,
Please Contact:

Product	Product number
• Biocrates® Bile Acids Kit (96)	9120052120813
• Biocrates® Bile Acids Kit (56)	9120052120905
• Biocrates® Bile Acids Kit (U)HPLC Column	9120052120868
• Biocrates® Bile Acids Kit (U)HPLC Precolumn	9120052120875
• Biocrates® Bile Acids Kit (U)HPLC Setup Box	9120052120882

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