C146-E280D

SHIMADZU
Excellence in Science

Selection Guide Metabolite Analysis

Metabolomics Product Portfolio

Expanding Metabolomics

Metabolomics refers to an array of techniques used to comprehensively detect and analyze various metabolites formed in vivo during biological activity. The qualitative and quantitative changes in metabolites reflect the ever-changing biological phenomena and are widely used for diagnosis, biomarker discovery, and drug discovery research. In recent years, metabolomics has been used in the food industry to improve taste and quality and to develop functional foods. It is also used in the biotechnology industry to improve fermentation and biofuel productivity.
Shimadzu supports the development and proliferation of metabolomics technologies by providing solutions combining mass spectrometers, databases, and software, which cover quantitative metabolomics, non-target analysis and multi-omics.

Medical Research

- Elucidation of physiological and pathological mechanisms
- Disease biomarker discovery
- Drug discovery support and toxicity evaluation

Plant and Food

- Analysis of flavor and fragrance components
- Functional evaluation
- Food authenticity and quality control

Biotechnology

- Improvement and optimization of fermentation process
- Biofuel productivity improvement

Contents

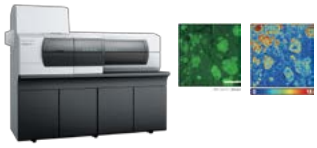
Product portfolio supports efficient and effective metabolomics from quantitative metabolomics to multi-omics analysis

Mass Spectrometers

GC-MS/MS

Q-TOF LC/MS

LC-MS/MS

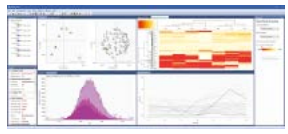


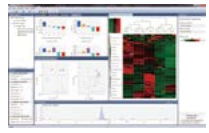
Imaging Mass Microscope

Database (Method Packages)

- Primary metabolites
- Metabolic enzymes
- D/L amino acids
- Cell culture profiling
- Lipid mediators
- Phospholipid profiling
- Short chain fatty acids
- Bile acids

Software





Multivariate Analysis

Visualization of Data

Provide the optimal products and workflow according to the objective

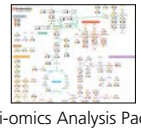
Wide Target Quantitative Metabolomics (pp. 4–5)

The combination of triple-quadrupole MS and database with a wide range of registered primary metabolite analysis conditions makes it possible to obtain large amounts of information efficiently, including simultaneous quantitative analysis of 475 compounds.

GCMS-TQ8050 NX/LCMS-8060

Traverse MS

LC/MS/MS Method Package for Primary Metabolites



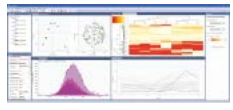
Multi-omics Analysis Package

Q-TOF LC/MS Based Non-target/Target Metabolomics (pp. 6–7)

The combination of highly sensitive and stable Q-TOF LC/MS and multivariate analysis software provides an effective workflow for non-target/target metabolomic analysis.

LCMS-9030

LC/MS/MS Method Package for Primary Metabolites and Cell Culture Profiling

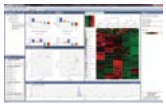


Signpost MS™

Lipidomics (pp. 10–11)

The combination of triple quadrupole LC-MS and database for lipid mediators/phospholipid profiling enables efficient lipid profiling.

LCMS-8060



Traverse MS

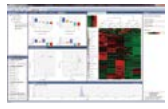
LC/MS/MS Method Package for Lipid Mediators and Phospholipid Profiling

Multi-omics Analysis Package

Multi-omics (pp. 10–17)

The combination of database and analysis software provides effective work-flows for multi-omics analysis such as metabolomics and lipidomics.

LCMS-8060



Traverse MS

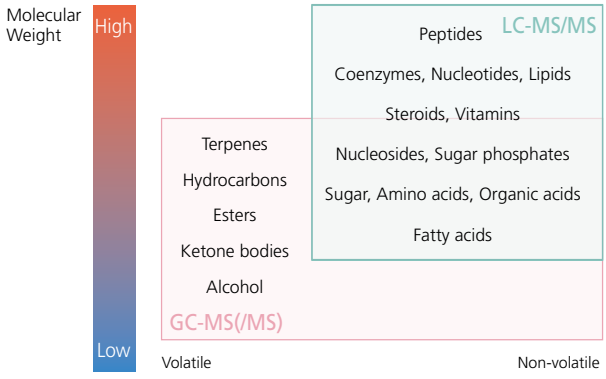
LC/MS/MS Method Package for Primary Metabolites and Lipid Mediators

Multi-omics Analysis Package

Wide Target Quantitative Metabolomics

TQ-MS with high sensitivity and ultra-fast performance and extensive metabolite database support reliable metabolomics.

Mass spectrometers are widely used in metabolomics with different techniques used depending on the object to be measured. Here, we introduce a "wide target quantitative metabolomics" approach that combines highly quantitative and stable triple-quadrupole LC-MS/MS and GC-MS(/MS) with databases for the primary metabolites analysis. This approach allows a wide range of metabolites to be analyzed simultaneously without the need to set complicated analysis conditions.



GC-MS(/MS)

LC-MS/MS

Instrument Features

GC-MS(/MS)
Permits comprehensive measurement of several hundred compounds in a single measurement
Standard measurement methods with excellent robustness
Low installation cost

LC-MS/MS
Simple measurement of specific metabolites (up to 100 compounds)
Quick measurement, including pretreatment
Measurement of high-molecular weight non-volatile metabolites is possible

Quantitative Metabolomics Using LC/MS/MS

LC-MS is the most widely used technique for metabolomics because many metabolites such as sugars, amino acids, and organic acids are hydrophilic compounds and can be measured by simple pretreatments. Combining Shimadzu's highly sensitive and ultra-fast LC-MS/MS with a method package for primary metabolites enables rapid and efficient quantitative metabolite analysis.

*For sample pretreatments, refer to *Pretreatment Procedure Handbook for Metabolites Analysis* (C146-E323).

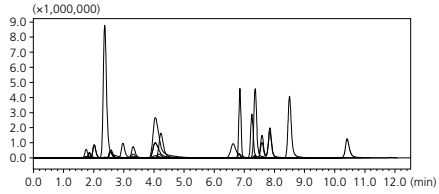
Target Metabolic Pathway	Method	Number of Registered Compounds
Glycolysis, pentose phosphate pathway, coenzyme, etc.	Ion pair method	55
Methylation cycle, urea cycle, TCA cycle, etc.	Non-ion pair method	97

This package includes two methods, permitting selection of the method that suits the target compounds and instrument environment. It should be noted that the PFPP column is used with the non-ion pair method.

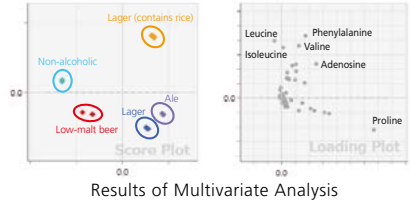
*Please refer to page 20 for more information on the compounds included in this method package.

■ Analysis example of five beer varieties

The Method Package for Primary Metabolites was used to perform simultaneous analysis (non-ion pairing method) of five types of commercially available beers. Based on the measurement results, principal component analysis and hierarchical cluster analysis were carried out, and it was confirmed that the difference between the five beer components indicates clusters according to their characteristics. This method can also be applied to the analysis and evaluation of a wide range of foods, including other alcoholic beverages.



MRM Chromatogram of Metabolites in Beer



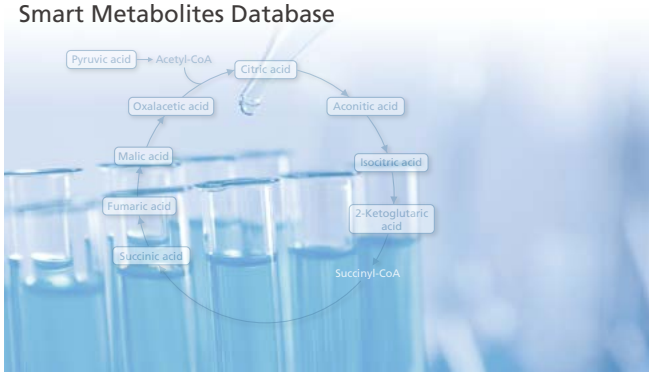
Results of Multivariate Analysis

GC-MS/MS Based Metabolomics

Metabolomics samples contain various metabolites and contaminants. Because GC-MS(/MS) offers excellent chromatographic resolution, high sensitivity and stable measurement, this technique is widely used in metabolomics. Shimadzu's Smart Metabolites Database offers a wide range of primary metabolite analysis conditions, enabling simultaneous analysis of 475 metabolites using MRM measurement and efficient acquisition of large amounts of information.

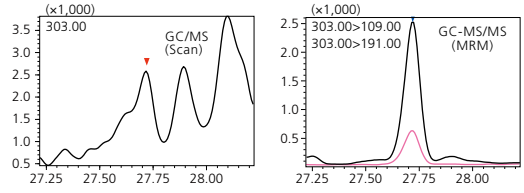
*For sample pretreatments, refer to *Pretreatment Procedure Handbook for Metabolites Analysis* (C146-E323).

Smart Metabolites Database



Registered Compounds	Derivative	Measurement	Number of Registered Compounds
Organic acids, fatty acids Amino acids, sugars, etc.	TMS*1	Scan	568
		MRM	475
Fatty acids	Methylation	Scan	50
		MRM	50
Amino acids	EZ:faast™ *2	Scan	33

*1 TMS indicates a trimethylsilyl derivative, and methylation represents the methyl ester derivative.
*2 EZ:faast is a product of Phenomenex Inc.
*3 Please refer to page 21 for more information on the compounds included in this method package.
This database is based on joint research with the Faculty of Medicine, Shimane University Graduate School of Medicine, Kobe University, and the Institute for Integrated Cell-Material Sciences, Kyoto University.



Comparison of Chromatograms of GC/MS and GC-MS/MS Analysis

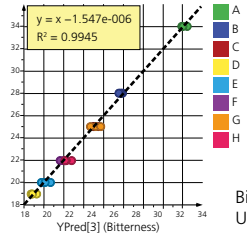
GC/MS/MS analysis is highly selective and less susceptible to contaminants. It has been confirmed that more accurate knowledge can be obtained in metabolomics by using GC-MS/MS.

■ Analysis example: construction of a regression model for a coffee sensory evaluation

Eight types of coffee beans were ground, roasted, and extracted under the same conditions and subjected to sensory evaluation. Metabolites were then extracted from each coffee bean and measured using GC-MS/MS. Using the results of the sensory evaluation as response variables and the processed values of the detected peak areas of metabolites as explanatory variables, a partial least squares (PLS) regression model of the relationship among these variables was constructed.

Results of Sensory Evaluation

Sample	A	B	C	D	E	F	G	H
Bitterness score	34	28	25	19	20	22	25	22



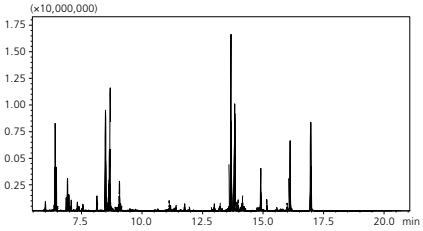
Bitterness Score Prediction Plot Using PLS Regression Model

Bitterness Score and Compounds with a Large Regression Coefficient

Compound Name	VIP	Regression Coefficient
Glycine-3TMS	1.648	0.047
Arabitol-5TMS	1.682	0.043
Mannitol-6TMS	1.783	0.042
Glucose-meto-5TMS	1.772	0.041
3-Phenyllactic acid-2TMS	1.591	0.037
Gluconic acid-6TMS	1.342	–0.033
Coniferyl aldehyde-meto-TMS	1.332	–0.033
Erythrulose-meto-3TMS	1.383	–0.034
Glyceraldehyde-meto-2TMS	1.578	–0.037
4-Hydroxybenzonic acid-2TMS	1.574	–0.037

Measurement Examples

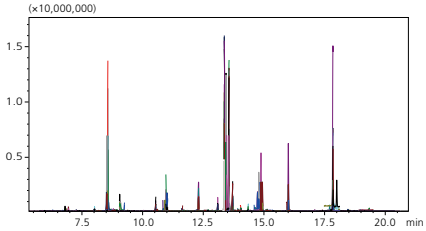
Human Standard Serum (TMS-derivatized MRM)



221 metabolites, including amino acids, organic acids, fatty acids, and sugars, were identified.

Reference: GC-MS Data Sheet No. 104 (LAAN-J-MS-E104)

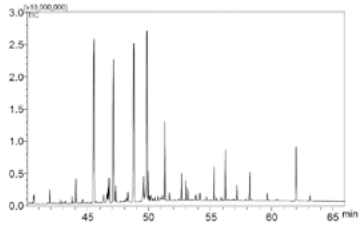
Mature Tomato Leaf (TMS-derivatized MRM)



170 metabolites, including glycolytic system and TCA cycle metabolites, were identified.

Reference: Technical Report (C146-E315)

Human ES Cells (Methylated)



19 kinds of fatty acids, including saturated fatty acids and unsaturated fatty acids, were identified.

Reference: Technical Report (C146-E243)

Data on the mature tomato leaf was provided by Dr. Yuji Sawada and Dr. Masami Yokota Hirai of the RIKEN Center for Sustainable Resource Science. This research was conducted by Japan's Council for Science, Technology and Innovation, Cross-ministerial Strategic Innovation Promotion Program (SIP) "Technologies for creating next-generation agriculture, forestry and fisheries" (administrative body: NARO Bio-oriented Technology Research Advancement Institution (BRAIN)).

Human ES cells were provided by Dr. Norio Nakatsuji and Dr. Kazuhiro Aiba of the Institute for Integrated Cell-Material Sciences, Kyoto University.
This achievement was obtained as a result of a project commissioned by the New Energy and Industrial Technology Development Organization (NEDO) to develop fundamental technology for enhancing industrial applications of human stem cells, as well as to develop fundamental assessment technology for their practical applications.

Q-TOF LC/MS Based Non-target/Target Metabolomics

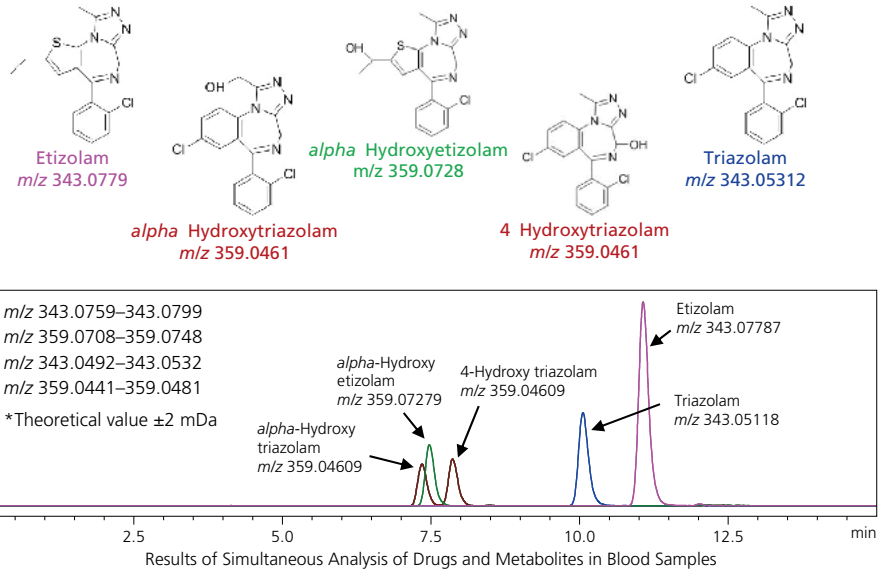
HR-MS with high sensitivity, accuracy and stability provides effective metabolomics workflows.

Combining technologies cultivated in the triple quad LCMS-8000 series with new TOF technologies, the Q-TOF LCMS-9030, under its product tag line "Effortless Performance", features not only superior sensitivity and resolution but also stable acquisition of high-mass accuracy data. Shimadzu also provides effective solutions for non-target analysis in metabolomics research.

Effortless Performance

Greater Accuracy
Better Sensitivity
Higher Resolution

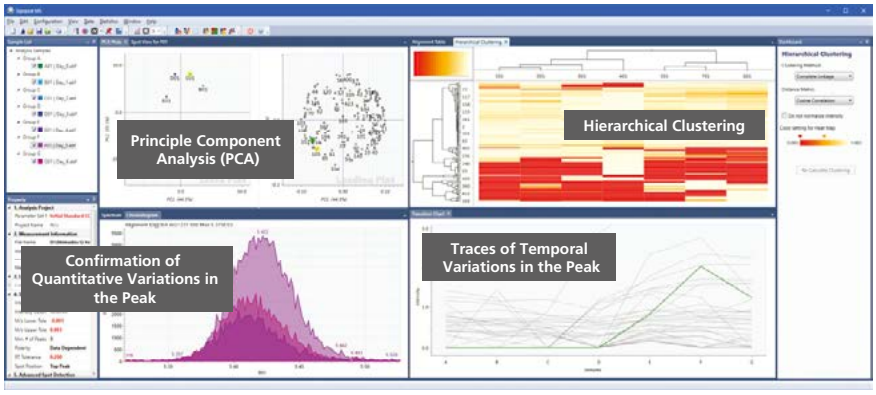
LCMS-9030 Quadrupole Time-of-Flight
Liquid Chromatograph Mass Spectrometer



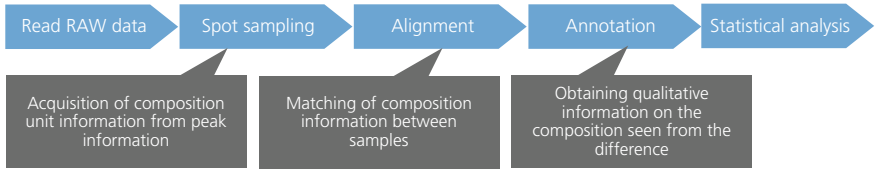
Results of Simultaneous Analysis of Drugs and Metabolites in Blood Samples

Multivariate Analysis Software: Signpost MS

Signpost MS is a multivariate analysis software that automatically picks up peaks from mass spectrometry data and assigns them in order to extract molecular (ion/fragment) information, which enables a comparison among samples. It is effective for analysis of non-targeted data acquisition and supports the data file formats of LC/MS/MS, Q-TOF LC/MS, GC/MS, and GC/MS/MS.



Data Processing Flow

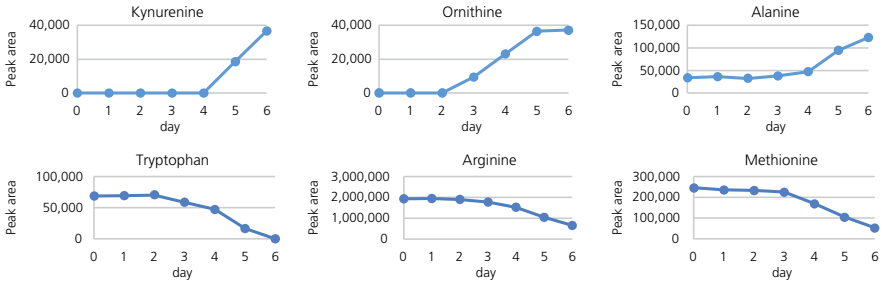


* Signpost MS is a product of Reifycs Inc.

■ Analysis example: Comprehensive cell culture profiling using the Q-TOF LC/MS

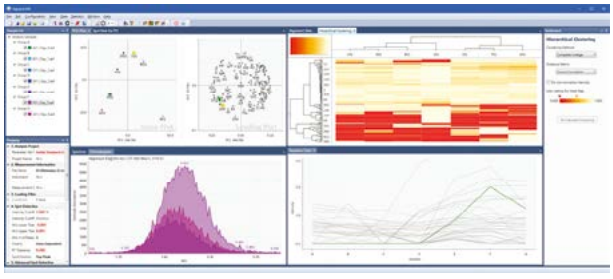
● Targeted Analysis Using SIM

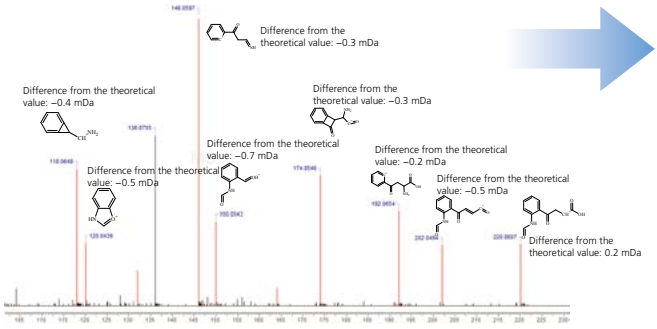
Sample	
Cell line	Feeder-free iPS cells 1231A3
Passage number	0P30
Seeding number	1.3 × 104 cells/well
Period	6 days
Medium	AK02N
Cell substrate	iMatrix (0.5 μg/cm²)

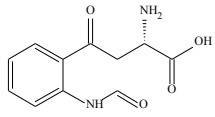


A SIM analysis of the culture supernatant using the analysis conditions listed in "LC/MS/MS Method Package for Cell Culture Profiling" resulted in 27 compounds, including amino acids and vitamins, being detected. It is evident that kynurenine, ornithine, and alanine increased as the cultivation time progressed, while there was a tendency for tryptophan, arginine, and methionine to decrease.

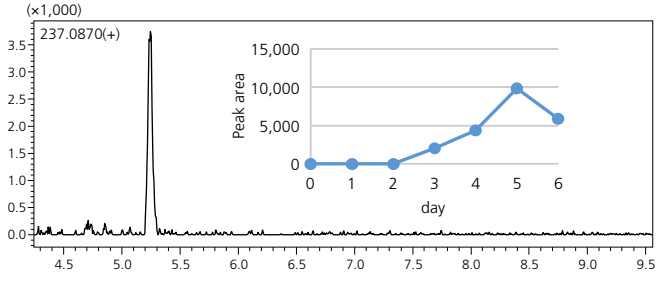
● Non-targeted Full Scan Analysis





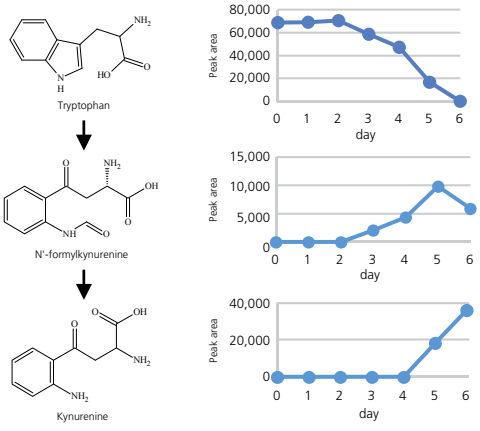


$C_{11}H_{12}N_2O_4$
(N′-formyl kynurenine)



A non-targeted analysis was implemented from the full scan analysis data in order to search for compounds other than the SIM targeted compounds for which quantitative fluctuations were observed. From the analysis results, it was confirmed that some of the compounds experienced quantitative fluctuations were not included in the SIM targets. As an example of compound estimation, an unknown metabolite with the retention time of 5.25 and 237.0870 *m/z* was estimated to be N′-formylkynurenine by database search and fragment peaks analysis.

● Analysis Result



N′-formylkynurenine is an intermediate metabolite of tryptophan and kynurenine in the kynurenine pathway. From the results for tryptophan and kynurenine, analyzed with SIM, and the results for N′-formylkynurenine from the non-targeted analysis, it is believed that tryptophan is taken into cells from the culture media in accordance with the cultivation process, reducing its concentration in the culture media. The metabolites of this, N′-formylkynurenine and kynurenine, increase in concentration in the culture media through secretion to the cell exterior. The decrease in N′-formylkynurenine on the sixth day is believed to be because the tryptophan within the culture media was depleted. Combining targeted SIM with non-targeted full scan analysis makes it possible to perform comprehensive cell culture profiling.

* The samples used for this data acquisition were cultures of iPS cells cultured at iPS PORTAL, Inc.

Imaging Metabolomics

MS-imaging technologies visualize vital phenomena

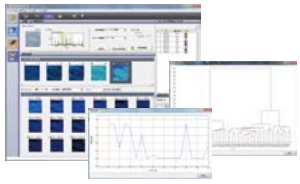
One limitation of conventional metabolomics is the loss of spatial distribution information due to the use of homogenized samples. Localization analysis, on the other hand, utilizes a slice of sample, enabling a more multi-faceted analysis.

Pretreatment Protocol (Example: Tissue)

1 Section preparation	• Fresh frozen section • Section thickness: About 10±5 µm • No fixing
2 Section mounting	• Use of a conductive glass slide
3 Imaging	• Capture optical microscope image
4 Matrix spray	• Deposition process by iMLayer • Spray process using air brush, etc.
5 Imaging mass spectrometry	• Superimposing image • Statistical analysis

Statistical Analysis Tools and iMLayer

HCA (Hierarchical Cluster Analysis)



Grouping by similarity of distribution images

ROI

Examining significant differences between regions of interest

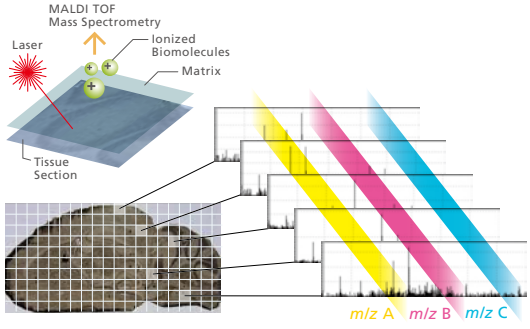
iMLayer

High-resolution imaging mass spectrometry by formation of fine crystals using vapor deposition

Tissue Imaging Overview

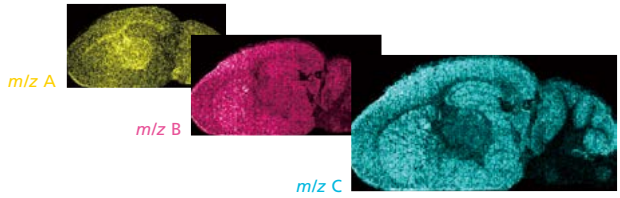
Mass Spectrometry at Multiple Points

Laser irradiation on a matrix-coated tissue section



Multi-Imaging

Visualization of molecular distribution based on signal intensities of specific ions



Imaging mass spectrometry involves using a mass spectrometer to measure biological molecules and metabolites directly while retaining information about their positions in a tissue sample. A 2D distribution map of each biological molecule is created based on the positional information obtained through measurements and the signal intensities of specific ions in mass spectra.

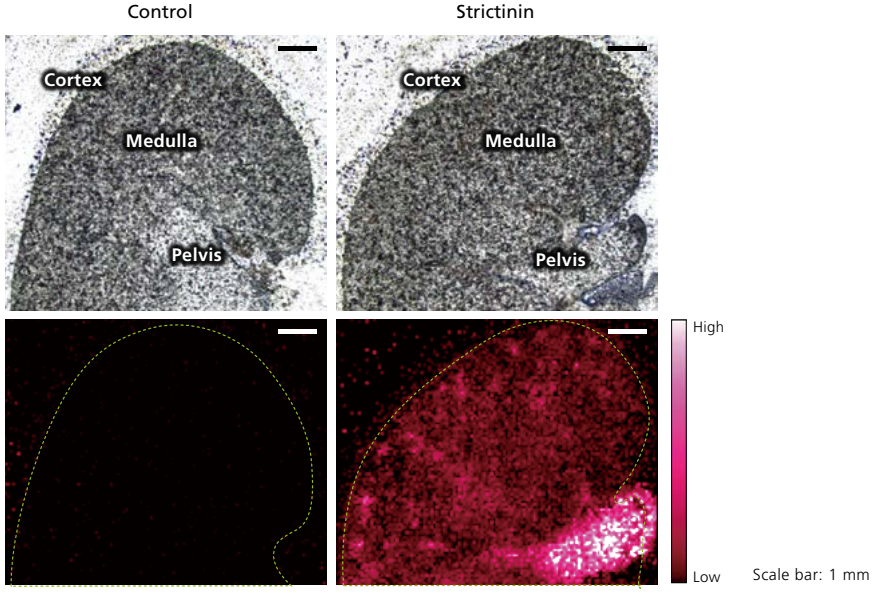
The iMScope *TRIO* is a system designed specifically for imaging mass spectrometry.
It is a hybrid microscope that combines an optical microscope and mass spectrometer to enable material structural analysis, which broadens possibilities in all fields of research.

MS-based Imaging Metabolomics

The combination of morphological information obtained by optical microscopy and functional analysis obtained by imaging mass spectrometry has found application in the field of metabolomics. In particular, obtaining information on the distribution of functional metabolites in living tissue, as well as the distribution of specific molecules in agricultural, food and crude drug products are key examples of imaging metabolomics applications.

■ Example: Analysis of Distribution of Functional Food Components

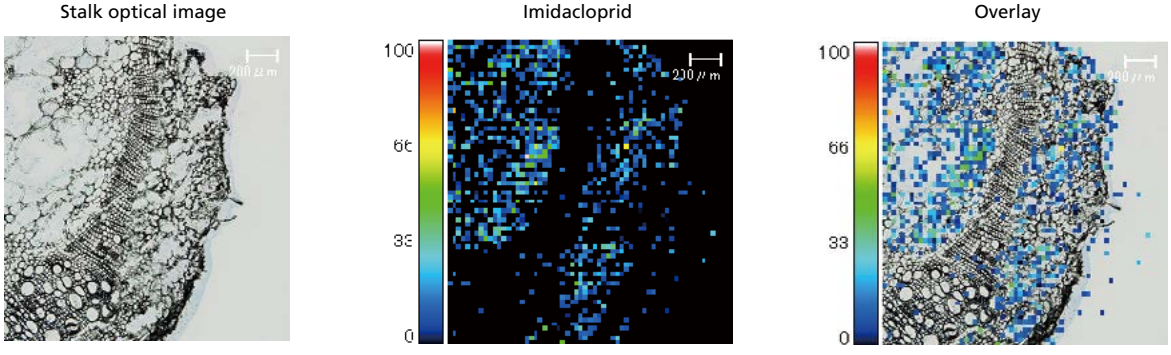
Strictinin, a polyphenol found in green tea, is attracting attention as a functional food constituent that is physiologically active in various ways. Accumulation of strictinin in the mice renal pelvis was demonstrated (oral administration).



This data was provided by Assistant Professor Daisuke Miura and Yoshinori Fujimura at Kyushu University.

■ Example: Analysis of Pesticide Distribution

Although neonicotinoid pesticides such as imidacloprid are frequently used as a substitute for organophosphorus pesticides due to their high permeability, some parts of the world have begun to restrict their use.
The distribution of imidacloprid in a tomato stem cross-section was measured in tomato plants after the pesticide was taken up by the tomato plant root. Considerable imidacloprid accumulation was observed in the cortical layer close to the surface and inside the xylem. Imaging techniques that enable observation of changes in distribution and quantity over time and by location can be used for research into topics such as the retention time of pesticides and optimization of insecticidal effect.



This data was provided by Associate Professor Shuichi Shimma at Osaka University.

Various Databases Containing "Ready-to-Use Methods"

Shimadzu provides comprehensive solutions for quantitative analysis

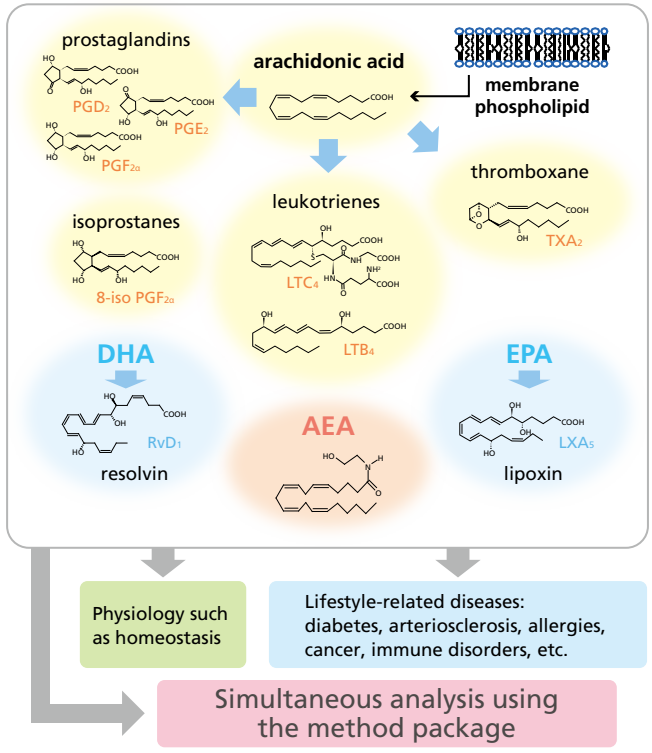
The various databases provided by Shimadzu contain "Ready-to-Use" methods and know-how. Analysis can be performed without performing complicated procedures, such as determining separation conditions for mass spectrometry or optimizing MS parameters for each compound, enabling efficient multi-component simultaneous analysis.

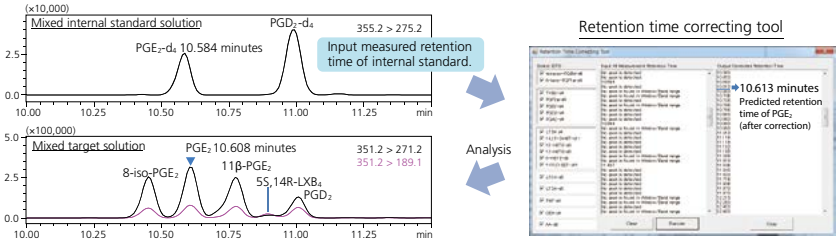
LC/MS/MS Method Package for Lipid Mediators

Lipid mediators (bioactive lipids) have important physiological functions and have been associated with allergies, thrombosis and lifestyle-related diseases. The LC/MS/MS Method Package for Lipid Mediators provides a simultaneous analysis method that encompasses 214 compounds, which include 196 compounds of lipid mediators derived from arachidonic acid cascade and 18 internal standard compounds. All components can be monitored in only 20 minutes.

Registered Compounds	Number of Registered Compounds
Arachidonic acid and its metabolites	100
EPA and its metabolites	26
DHA and its metabolites	23
Ethanol amides	11
Other fatty acid metabolites	36
Total	196

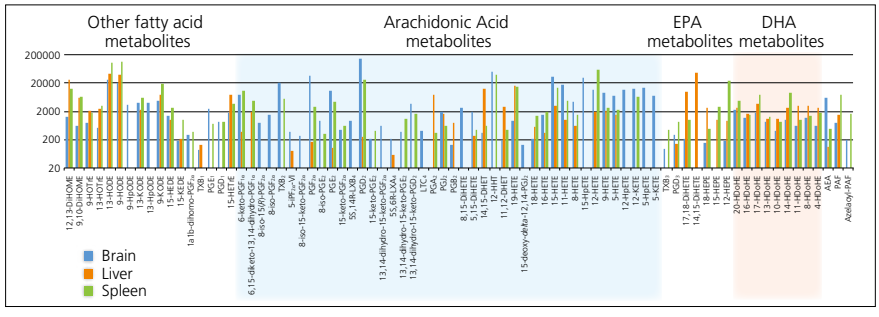
* Please refer to page 22 for more information on the compounds included in this method package.





■ Retention Time Correcting Tool Supports Identification of Isomers

The Retention Time Correcting Tool available in this method package simplifies retention time correction, enabling precise identification of isomers that cannot be distinguished by MRM. The 196 compounds are divided into 18 groups based on their properties, and internal standard samples have been chosen for each group, making it possible to correct for quantitation errors that may arise, such as during solid phase extraction.



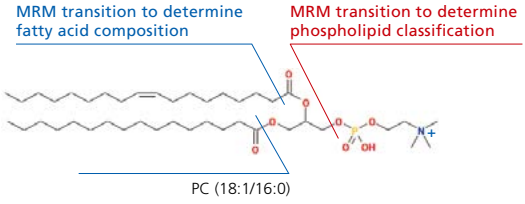
■ Analysis example: Lipid mediator profiling for a brain, liver and spleen tissue from a mouse

After lipid compounds were extracted, the extracted lipid mediators purified by a solid phase extraction were analyzed simultaneously. It was confirmed that a wide dynamic range profiling is possible at a low-concentration region.

LC/MS/MS MRM Library for Phospholipid Profiling

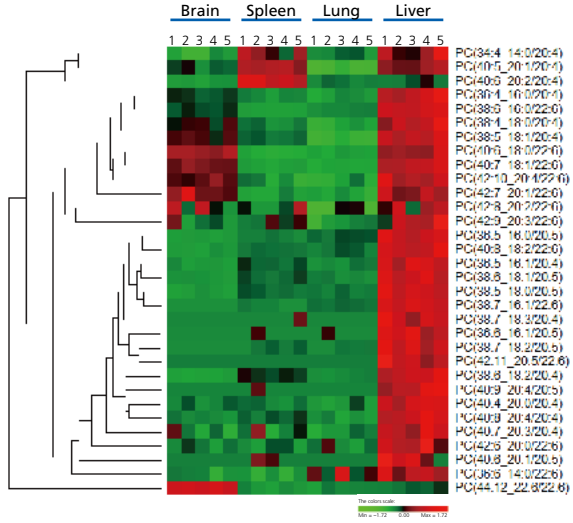
This MRM library includes two methods: one for phospholipid classification by comprehensive analysis of the main phospholipids in biological samples, and one for fatty acid composition determination created using analytical results obtained with the classification method. The library targets phospholipids containing C14 to C22 fatty acids, and includes MRM transitions for up to 867 components. This library enables performing phospholipid profiling by conducting an initial analysis with a phospholipid classification method. This is followed by creating a method for fatty acid composition determination based on the phospholipid peak detected in the first analysis, and subsequently using this method to perform a second analysis to determine fatty acid composition.

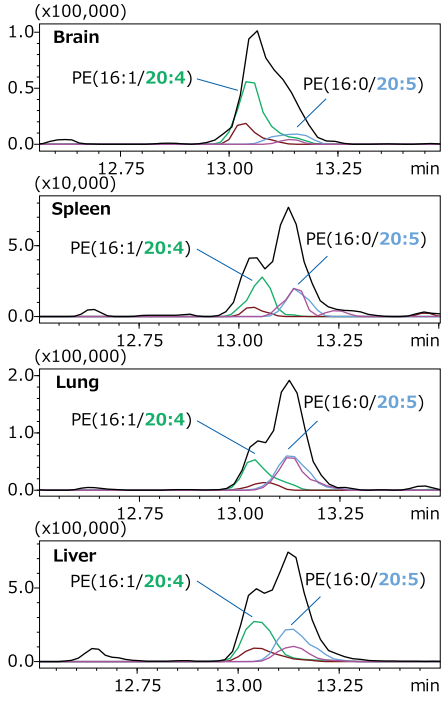
	Number of Double Bonds					
Carbon Number	C14:0	C14:1				
	C16:0	C16:1				
	C18:0	C18:1	C18:2	C18:3		
	C20:0	C20:1	C20:2	C20:3	C20:4	C20:5
	C22:0	C22:1	C22:6			



■ Analysis example: Phospholipid analysis in four types of mouse tissue —Cluster analysis of PUFA-containing PCs—

Using this library, a total of 225 phospholipid components were identified by analysis of four different tissue extracts. The results of a cluster analysis of phosphatidylcholine (PC) containing highly unsaturated fatty acids (PUFA) are shown below. It was confirmed that PC is mainly found in the liver, and 2 DHA bound components (bottom line) are mainly found in the brain.

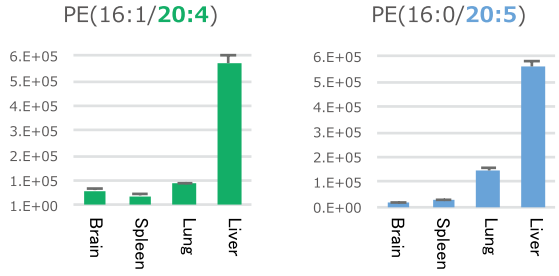




Polarity Switching MRM
PE(36:5) 738>597(pos)
PE(16:1/20:4) 736>253(neg) ×3.0 PE(16:0/20:5) 736>255(neg) ×3.0
PE(16:1/20:4) 736>303(neg) ×3.0 PE(16:0/20:5) 736>301(neg) ×3.0

■ Analysis example: Phospholipid analysis in four types of mouse tissues —Quantitative profiling of PE (36:5)—

PE (16:1/20:4) and PE (16:0/20:5) are identical in molecular weight and cannot be distinguished by precise mass spectrometry alone. However, they were separated and detected as components with different retention times as a result of simultaneous monitoring with high-speed polarity switching of 5 msec. It is also possible to compare the ratio of fatty acids in phospholipid components between samples. A comparison between PE (16:1/20:4) and PE (16:0/20:5) organs is shown in the graph below.

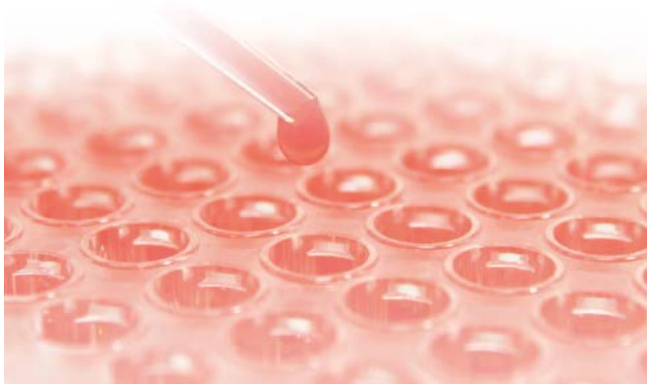


Comparison of the Ratio of the Amount of Fatty Acids Contained in Phospholipid Components between Samples

*Lipid extracts from mouse tissues were provided by Prof. Suzumi Tokuoka and Prof. Yoshihiro Kita of Advanced Lipidomics Research, Department of Lipidomics, The University of Tokyo.

LC/MS/MS Method Package for Cell Culture Profiling

The LC/MS/MS Method Package for Cell Culture Profiling contains the conditions for the simultaneous analysis of 95 major culture medium components and metabolites secreted by cells. It can provide useful knowledge for improving the production of target substances and optimizing culture conditions in bioproduction by measuring the time course of the culture supernatant.



Features

- Provides simultaneous analysis conditions for 95 components
- Enables simultaneous analysis in 17 minutes
- Enables simultaneous analysis of high-concentration and trace components

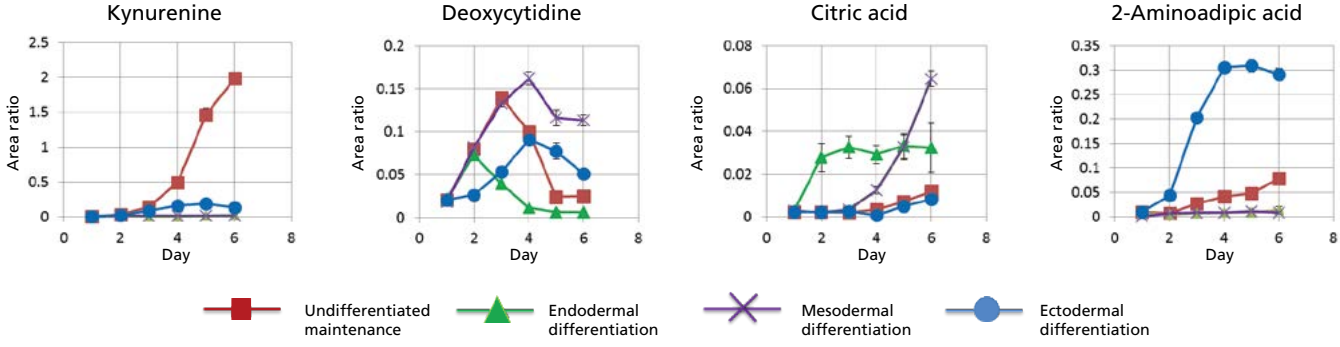
Registered Compounds	Number of Registered Compounds
Amino acids and derivatives	38
Nucleic acid-related compounds	18
Vitamins	17
Sugars	5
Others	17

*Please refer to page 20 for more information on the compounds included in this method package.

■ Analysis example: Comparison of time course of culture supernatant components of undifferentiated iPS cells and differentiation-induced cells

One of the characteristics of human iPS cells is that they remain undifferentiated. Cell invasive methods such as gene expression analysis are common for this characterization. By analyzing the components of the culture supernatant, we examined whether we could distinguish the undifferentiated state from the differentiated state of iPS cells without destroying the cells.

As shown below, compounds showing characteristic temporal changes in the differentiation state of each cell were identified. Multicomponent analysis of the culture supernatant suggested that the undifferentiated state and the differentiated state of iPS cells could be distinguished.

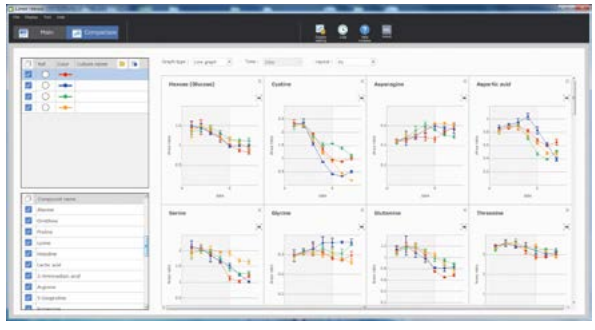


C2MAP: Cell Culture Media Analysis Platform

*Cell culture profiling method files are included in the control software.

Features

- Automated process from pretreatment to measurement for the culture supernatant analysis.
- Temporal changes in the components can be displayed as trend graphs.



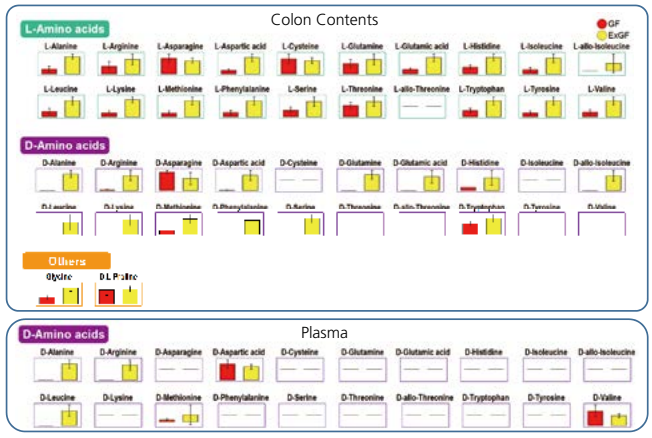
LC/MS/MS Method Package for D/L Amino Acids

With conventional chiral amino acid analysis, it is necessary to perform derivatization or use very long run times. With this method package, derivatization is not necessary, and high-sensitivity analysis can be performed in a short period of time, bringing efficiency to the chiral separations.

List of Registered Amino Acids		
D/L-Alanine	D/L-Histidine	D/L-Serine
D/L-Arginine	D/L-Isoleucine	D/L-Threonine
D/L-Asparagine	D/L-allo-Isoleucine	D/L-allo-Threonine
D/L-Aspartic acid	D/L-Leucine	D/L-Tryptophane
D/L-Cysteine	D/L-Lysine	D/L-Tyrosine
D/L-Glutamine	D/L-Methionine	D/L-Valine
D/L-Glutamic acid	D/L-Phenylalanine	
Glycine	DL-Proline	

■ Analysis example: D/L amino acid analysis in colon contents and plasma

A comprehensive analysis of chiral amino acids in mouse colon contents and plasma was conducted to investigate D-amino acids produced by intestinal flora. Concentrations of 12 D-amino acids were significantly higher in normal colonized mice (Ex-GF) than in sterile mice (GF), which indicates that these D-amino acids are produced by the intestinal flora.

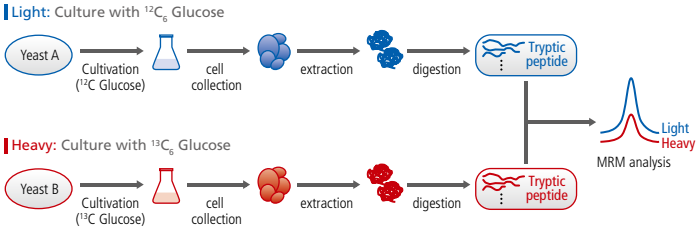


* The analysis method of this method package was developed based on the research results of the Fukusaki Lab, Division of Science and Biotechnology, Graduate School of Engineering, Osaka University.
Reference: Nakano, Y., Konya, Y., Taniguchi, M., Fukusaki, E., *Journal of Bioscience and Bioengineering*, 123, 134–138 (2016)

LC/MS/MS MRM Library for Metabolic Enzymes in Yeast

This product provides a library consisting of 3,584 MRM transitions, including stable isotopes. It covers all 498 trypsin digested peptides of 228 types of enzymes derived from budding yeast, which is used for the production of bioethanol and other materials, or as a model organism for basic research. This library enables a variety of enzyme measurements, including those related to the major metabolic pathways of glycolysis, the TCA cycle, the pentose phosphate cycle, and amino acid metabolism.

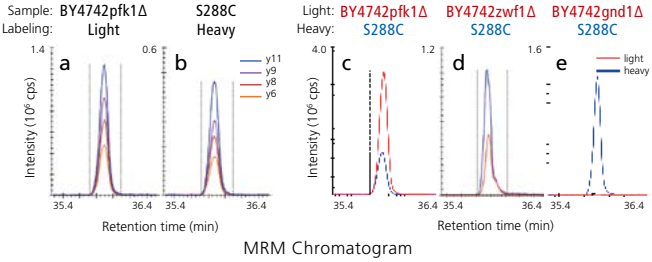
*Please refer to page 23 for more information on the compounds included in this library.



Yeast Metabolic Enzyme MRM Measurement Workflow

■ Analysis example: MRM analysis of Gnd1p trypsin digested peptides in gene-disrupted strains

Shown below are representative chromatograms for a BY4742pfk1Δ strain (light) grown with unlabeled glucose (a), and a S288C strain (heavy) grown with ^{13}C-labeled glucose (b). Additionally, TIC chromatograms of Gnd1p in gene-disrupted strains are shown in (c), (d), and (e). In GND1 disrupted strains, Gnd1p was not detected, whereas in PFK1 disrupted strains, large numbers of Gnd1p were detected.



MRM Chromatogram

LC/MS/MS Method Package for Short Chain Fatty Acids

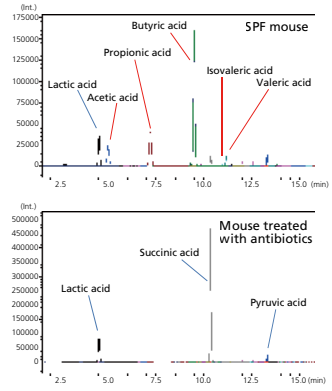
As the short-chain fatty acids produced by intestinal bacteria, acetic acid, propionic acid, and butyric acid are well known, and it has been reported that there are some connections between them and lifestyle-related diseases such as obesity and diabetes. Generally speaking, short-chain fatty acids are highly volatile and highly hydrophilic. This makes it difficult to perform LC/MS analysis using a conventional reversed phase system. For that reason, this method package targets short-chain fatty acids (C2 to C5) that have been derivatized using 3-nitrophenylhydrazine (3-NPH). After setting MRM transitions, it can be used for the simultaneous analysis of 22 components, including organic acids related to the central metabolic pathways.

Short-Chain Fatty Acids
Acetic acid
Propionic acid
Butyric acid
Isobutyric acid
Valeric acid
Isovaleric acid

Organic Acids	
2-oxobutyric acid	Glycolic acid
2-hydroxyglutaric acid	Succinic acid
α-ketoglutaric acid	Lactic acid
β-hydroxybutyric acid	Pyruvic acid
Isocitric acid	Fumaric acid
Oxaloacetic acid	Maleic acid
Citric acid	Malonic acid
Glyoxylic acid	Malic acid

■ Analysis example: Analysis of short chain fatty acids and organic acids in mouse fecal samples

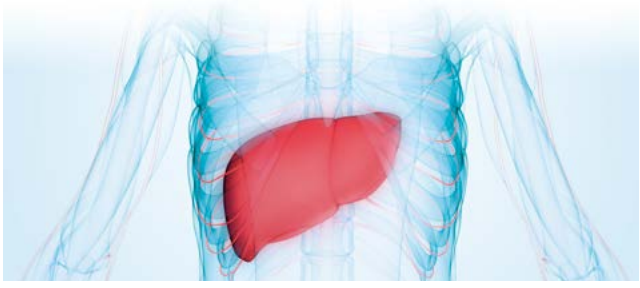
Characteristic changes in the amounts of short chain fatty acids and organic acids in the samples were observed under conditions in which the intestinal microbiota was decreased by sterile feeding and antibiotics.



MRM Chromatogram

LC/MS/MS Method Package for Bile Acids

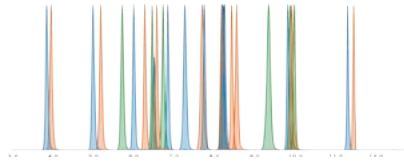
Bile acids are primarily produced through the breakdown of cholesterol in the liver. Primary bile acids can then be conjugated with taurine or glycine and/or converted into secondary bile acids by gut bacteria. Their role as digestive surfactants to promote the absorption of lipids is well-known, but they also act as hormones in the regulation of various metabolic pathways. This method package contains the analytical conditions for 28 major components of primary and secondary bile acids and their conjugates



*Please refer to page 23 for more information on the compounds included in this method package.

■ Analysis example: Analysis of 28 bile acids (standard sample)

The following is an example of the simultaneous analysis of 28 components including primary and secondary bile acids and their taurine and glycine conjugates. It was confirmed that the separation and identification of each component could be performed in a 17-minute run time.



MRM Chromatogram

List of Database

GC/MS, GC-MS/MS Method Package for Metabolites

Description	Flyer code
Smart Metabolites Database	C146-E277

LC/MS/MS Method Packages

Description	Flyer code
Primary Metabolites	C146-E227
Short Chain Fatty Acid	C146-E355
Lipid Mediators	C146-E381
D/L Amino Acids	C146-E336
Cell Culture Profiling	C146-E279
Bile Acids	C146-E386

LC/MS/MS MRM Library

Description	Flyer code
Metabolic Enzymes in Yeast	C146-E275
Phospholipid Profiling	C146-E314

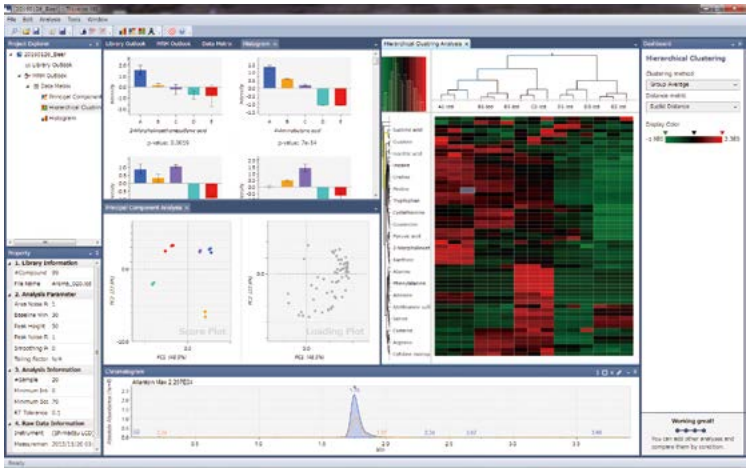
Software Supports Efficient and Effective Data Analysis

The software compatible with MS and database enables suitable workflows according to various demands.

The amount of information obtained from the omics approach to comprehensively analyze various compounds in living organisms, including metabolomics, is enormous, so it is essential to have software that can analyze data efficiently and effectively. Shimadzu provides a variety of data analysis software to support efficient data processing, multivariate analysis, and data visualization of multi-specimen MS data.
Each software and method packages are compatible. For example, data acquired using Method Package for Primary Metabolites can be statistically analyzed by Traverse MS, and further visualized on a metabolic map by using Multi-omics Analysis Package. A series of analysis tasks can be performed efficiently.

Traverse MS

Traverse MS data analysis software is intended for high-speed processing of MRM data acquired with Shimadzu triple quadrupole MS systems in the field of targeted metabolomics. Using multiple samples and multiple components, the software is able to create graphical and statistical analysis for metabolic pathway analysis.



Features

- Displays multiple chromatograms in a single window
- Peak identification algorithm specialized for MRM data
- Graphing area values (and area ratios) for multiple samples
- Statistical analysis based on principal component analysis and hierarchical cluster analysis
- Supports metabolic pathway analysis

*Traverse MS is a product of Reifycs Inc.

LabSolutions Insight

Mass spectrometry laboratories can acquire thousands of chromatograms per day. However, it takes a huge amount of time to confirm and analyze the data. LabSolutions Insight makes it easier and more efficient to analyze multiple samples.

Features

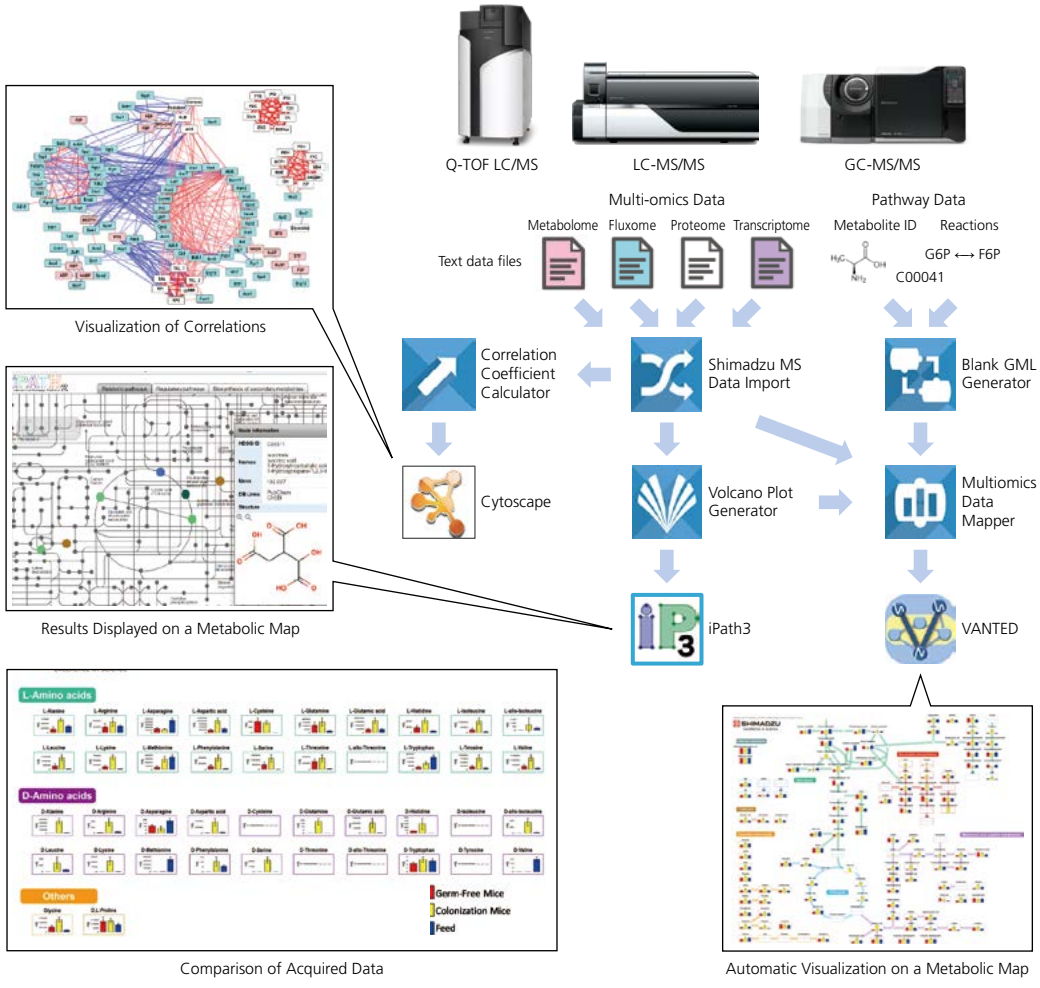
- Improved efficiency of multi-analyte quantitation and optimize workflow
- Significantly reduced time of multi-analyte quantitation (various flagging and peak waveform processing)
- Flexible work styles

■ Using LabSolutions Insight in combination with databases and method packages

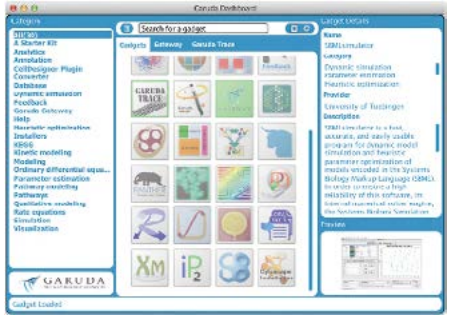
When used in combination with databases or application-specific method packages, the entire analytical process, from measurement to data analysis, can be performed easily without having to optimize analytical conditions.

Multi-omics Analysis Package

The Multi-omics Analysis Package, developed for metabolic engineering applications, provides the ability to automatically generate metabolic maps and perform a variety of data analysis for the vast data generated in fields like metabolomics, proteomics and flux analysis. It offers a powerful platform to support drug discovery, bioengineering and other life science research applications.



The Multi-omics Analysis Package is based on software tools (called gadgets) that have been released on the GARUDA™ platform — an open research platform, developed by the GARUDA Alliance led by The Systems Biology Institute, Japan (SBI).



http://www.garuda-alliance.org/

POWERED BY GARUDA

Data Analysis Tools Used in the Multi-omics Analysis Package

Volcano Plot

A tool that combines a t-test (statistically significant difference) and a fold-change (Example: Difference in mean value such as 2 times or 1/2) to visualize the differences between the two groups. The Volcano Plot gadget developed by Shimadzu is included in the package.

VANTED

Tool maintained at University of Konstanz, Germany, for visualization and analysis of networks across different data sets. (GARUDA support was developed at Monash University)

iPath

Data analysis tool developed by the European Molecular Biology Laboratory that can be used for visualization of diverse metabolic pathways or data mapping and customization.

Cytoscape

Bioinformatics tool developed by the Cytoscape Consortium, used to visualize metabolic pathways, to integrate gene expression profiles with related data, and so on. It is especially useful for analyzing networks and visualizing correlations.

■ Analysis example: Changes in metabolite levels in a cell culture medium over time

Changes in metabolite levels in a cell culture medium over time were measured using a GC-MS system. Acquired data were analyzed using the Multi-omics Analysis Package and metabolite changes were visualized by displaying them on a metabolic map.

Experiment Protocol

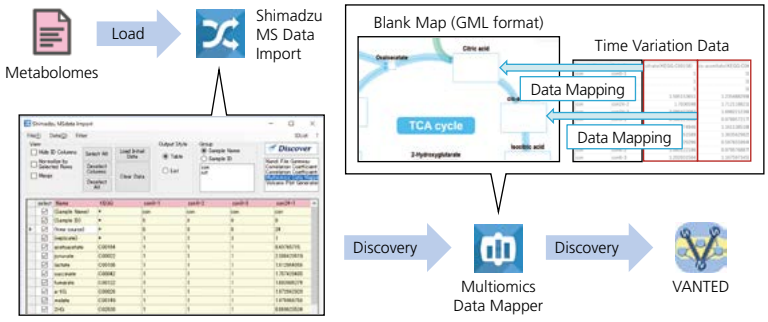
Culture MCF-7 cell line for 15 hours.

Replace culture medium with medium spiked with rotenone.

Collect culture medium after 0, 3, 6, 9, and 24 hours.

Use GC-MS system to measure metabolite quantities.

Analyze data using Multi-omics Analysis Package.



GC-MS/MS

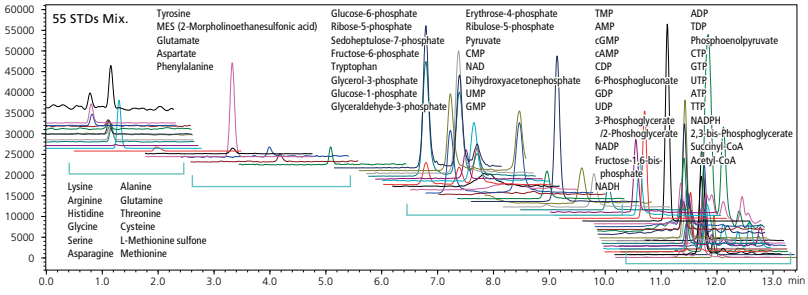
Advanced Solutions

Shimadzu pursues technological development and progress in metabolomics research through collaborative works.

Through joint research with external organizations, Shimadzu has been developing various technologies, products and applications for metabolomics. Shown below are examples of this joint research.

Parameter-free Peak Picking Technology Using AI

As the performance of mass spectrometers has improved, the amount of data that can be obtained has become enormous. Especially in metabolomics, the number of detected peaks is large, so picking detected peaks is a bottleneck in the analysis. In order to make analysis more efficient and improve the accuracy of data analysis, we are working with FUJITSU to develop a parameter-free peak picking method for chromatograms using AI technology.

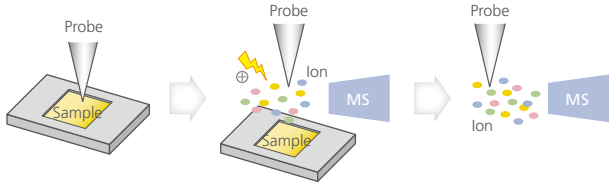


Direct Probe Ionization Mass Spectrometer

The DPiMS™-8060 uses a probe electrospray ionization technique to remove a very small amount of liquid from a sample that has been added to a sample plate. It performs mass spectrometry by ionizing the liquid using the probe, and introducing the liquid into the MS section. The micro-sampling by using the probe enables real-time monitoring of the change over time on the sample plate because it is used for analysis without changing the state of the sample.

DPiMS-8060

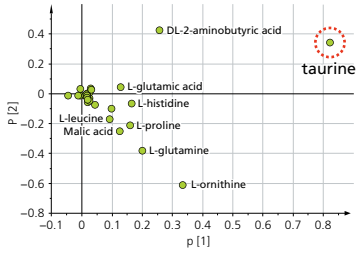
PESI (Probe Electrospray Ionization)-MS



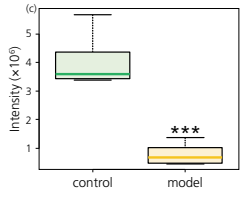
Sampling　Ionization　Mass spectrometry

■ Analysis example: Metabolomic analysis performed easily by DPiMS-8060

In this example, transition information for metabolites (26 components) such as amino acids, organic acids, and sugars was used for metabolomic analysis of mouse livers. Metabolomes can be analyzed easily by customizing some of the analytical conditions in the LCMS primary metabolite method package. The DPiMS-8060 system was used to measure the principal components in the model mouse group with acute liver damage induced by carbon tetrachloride and the control group. A significant difference was observed between the model and control groups, based on the significant contribution of taurine to the separation of groups in the PCA loading plot. The difference was verified with a box-whisker plot.



PCA Loading Plot



Box-whisker Plot of Taurine

* This data was obtained from joint research with Associate Professor Kei Zaitsu from Nagoya University Graduate School of Medicine.

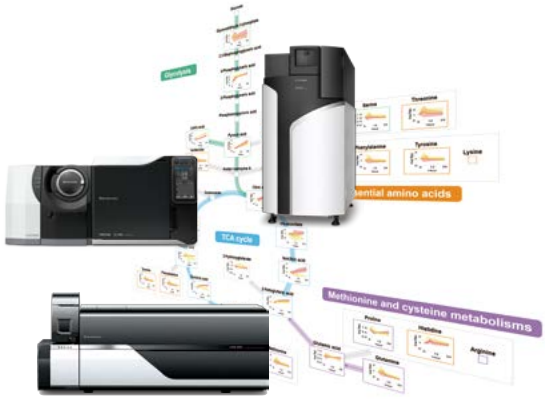
Joint Research Example:
Osaka University Shimadzu Analytical Innovation Research Laboratory

Collaboration and joint research are essential for technology development. As a base for external collaboration, this laboratory is putting efforts into research and development of preprocessing, analysis, data analysis methods and applications in metabolomics. In addition, the laboratory aims to further develop metabolomics by holding seminars, providing analysis consultations, and performing sample analysis.

Products Developed through Joint Research

Nexera™ UC
Supercritical Fluid Extraction/Chromatograph System

LC/MS/MS Method Package for D/L Amino Acids



LC/MS, GC/MS Data Analysis Software
Multi-omics Analysis Package

Articles on Metabolomics

In addition to this brochure, a list of articles related to metabolomics are posted on our website.

https://www.shimadzu.com/an/industry/pharmaceuticallifescience/n9j25k00000nga0n.html

List of Metabolites Measurable by LC-MS/MS

Grouping	Compound	PFPP	I.P.	C.C.
Glycolytic Pathway	2,3-Bisphosphoglyceric acid	–	○	–
	3-Phosphoglyceric acid (2-Phosphoglyceric acid)	–	○	–
	Dihydroxyacetone phosphate	–	○	–
	Fructose 1,6-bisphosphate	–	○	–
	Glucose 1-phosphate	–	○	–
	Glucose 6-phosphate	–	○	–
	Glycerol 3-phosphate	–	○	–
	Lactic acid	○	–	○
	Phosphoenolpyruvic acid	–	○	–
	Pyruvic acid	○	○	○
Pentose · Phosphate Pathway	6-Phosphogluconic acid	–	○	–
	Erythrose 4-phosphate	–	○	–
	Fructose 6-phosphate	–	○	–
	Glyceraldehyde 3-phosphate	–	○	–
	Ribose 5-phosphate	–	○	–
	Ribulose 5-phosphate	–	○	–
	Sedoheptulose 7-phosphate	–	○	–
TCA Cycle	2-Ketoglutaric acid	○	–	–
	Acetyl coenzyme A	–	○	–
	Aconitic acid	○	–	–
	Citric acid	○	–	○
	Fumaric acid	○	–	○
	Isocitric acid	○	–	○
	Malic acid	○	–	○
	Succinic acid	○	–	○
	Succinyl coenzyme A	–	○	–
Amino Acid-Related Metabolites	2-Aminoadipic acid	–	–	○
	2-Aminobutyric acid	○	–	–
	4-Aminobutyric acid	○	–	○
	4-Hydroxyproline	○	–	○
	5-Oxoproline	–	–	○
	Alanine	○	○	○
	Arginine	○	○	○
	Asparagine	○	○	○
	Aspartic acid	○	○	○
	Asymmetric dimethylarginine	○	–	–
	Citrulline	○	–	○
	Cystathionine	○	–	○
	Cysteine	○	○	○
	Cystine	○	–	○
	Dimethylglycine	○	–	–
	Glutamic acid	○	○	○
	Glutamine	○	○	○
	Glycine	○	○	○
	Histidine	○	○	○
	Homocysteine	○	–	–
	Homocystine	○	–	–
	Isoleucine	○	–	○
	Kynurenine	○	–	○
	Leucine	○	–	○
	Lysine	○	○	○
	Methionine	○	○	○
	Methionine sulfoxide	○	–	○
	N-Acetylasparatic acid	–	–	○
	N-Acetylcysteine	–	–	○

Grouping	Compound	PFPP	I.P.	C.C.
Amino Acid-Related Metabolites	Ornitine	○	–	○
	Phenylalanine	○	○	○
	Pipecolic acid	–	–	○
	Proline	○	–	○
	Serine	○	○	○
	Symmetric dimethylarginine	○	–	–
	Threonine	○	○	○
	Tryptophan	○	○	○
	Tyrosine	○	○	○
	Valine	○	–	○
Nucleic Acid-Related Compounds	Adenine	○	–	○
	Adenosine	○	–	○
	Adenosine 3′,5′-cyclic monophosphate	○	○	–
	Adenosine diphosphate	–	○	–
	Adenosine monophosphate	○	○	○
	Adenosine triphosphate	–	○	–
	Cytidine	○	–	○
	Cytidine 3′,5′-cyclic monophosphate	○	–	–
	Cytidine diphosphate	–	○	–
	Cytidine monophosphate	○	○	○
	Cytidine triphosphate	–	○	–
	Cytosine	○	–	–
	Deoxycytidine	–	–	○
	Guanine	○	–	○
	Guanosine	○	–	○
	Guanosine 3′,5′-cyclic monophosphate	○	○	–
	Guanosine diphosphate	–	○	–
	Guanosine monophosphate	○	○	○
	Guanosine triphosphate	–	○	–
	Hypoxanthine	○	–	○
	Inosine	○	–	○
	Thymidine	○	–	○
	Thymidine diphosphate	–	○	–
	Thymidine monophosphate	○	○	–
	Thymidine triphosphate	–	○	–
	Thymine	○	–	○
	Uracil	○	–	○
	Uric acid	○	–	○
	Uridine	○	–	○
	Uridine diphosphate	–	○	–
	Uridine monophosphate	–	○	–
	Uridine triphosphate	–	○	–
	Xanthine	○	–	○
	Xanthosine	–	–	○
Sugars	Gluconic acid	–	–	○
	Glucosamine	–	–	○
	Hexose (Glucose)	–	–	○
	Sucrose	–	–	○
	Threonic acid	–	–	○
Coenzymes	FAD	○	–	–
	FMN	○	–	–
	NAD	○	○	–
	NADH	–	○	–
	NADP	–	○	–
	NADPH	–	○	–

PFPP: PFPP column method, I.P.: Ion-pairing reagent method, C.C.: Cell culture profiling method

LC/MS/MS Method Package for Primary Metabolites and the LC/MS/MS Method Package for Cell Culture Profiling enable the measurement of 166 metabolic compounds, including the metabolites shown above, vitamins and di/tripeptides.
LC/MS/MS Method Package for Lipid Mediators enables the measurement of 158 lipid mediators.

List of Metabolites Measurable by GC-MS(/MS)

Grouping	Compound
Glycolytic Pathway	2-Phosphoglyceric acid
	3-Phosphoglyceric acid
	Dihydroxyacetone phosphate
	Fructose 6-phosphate
	Glucose
	Glucose 6-phosphate
	Glyceraldehyde 3-phosphate
	Glycerol 3-phosphate
	Lactic acid
	Phosphoenolpyruvic acid
Pentose · Phosphate Pathway	6-Phosphogluconic acid
	Erythrose 4-phosphate
	Gluconic acid
	Glucono-1,5-lactone
	Glyceraldehyde
	Glyceric acid
	Ribose
	Ribose 5-phosphate
	Ribulose 5-phosphate
	Sedoheptulose 7-phosphate
TCA Cycle	2-Ketoglutaric acid
	Aconitic acid
	Citric acid
	Fumaric acid
	Isocitric acid
	Malic acid
	Oxalacetic acid
	Pyruvic acid
	Succinic acid
Amino Acid	Alanine
	Asparagine
	Aspartic acid
	Cysteine
	Cystine
	Glutamic acid
	Glutamine
	Glycine
	Histidine
	Isoleucine
	Leucine
	Lysine
	Methionine
	Phenylalanine
	Proline
	Serine
	Threonine
	Tryptophan
	Tyrosine
	Valine
Urea Cycle	Arginine
	Citrulline
	Ornithine
	Urea

Grouping	Compound
Polyamine	Cadaverine
	Putrescine
	Spermidine
	Spermine
Nucleic Acid-Related Compounds	2′-Deoxyuridine
	5′-Methylthioadenosine
	7-Methylguanine
	Adenine
	Adenosine
	Adenosine 3′,5′-cyclic monophosphate
	Adenosine monophosphate
	Allantoin
	Cytidine
	Cytosine
	Guanine
	Guanosine
	Hypoxanthine
	Inosine
	Inosine monophosphate
	Orotic acid
	Paraxanthine
	Thymidine
	Thymidine monophosphate
	Thymine
	Uracil
	Uric acid
	Uridine
	Uridine monophosphate
	Xanthine
	Xanthosine
	Xanthosine monophosphate
Sugars	Allose
	Arabinose
	Erythrulose
	Fructose
	Fructose 1-phosphate
	Fucose
	Galactose
	Isomaltose
	Lactose
	Lyxose
	Maltose
	Mannose
	Mannose 6-phosphate
	Psicose
	Rhamnose
	Sorbose
	Sucrose
	Tagatose
	Trehalose
	Trehalose 6-phosphate
	Xylose
	Xylulose

Grouping	Compound
Amino Acid-Related Metabolites	2-Aminoadipic acid
	3-Aminoglutaric acid
	3-Aminopropanoic acid
	3-Hydroxyanthranilic acid
	3-Hydroxy-kynurenine
	3-Methyl-2-oxovaleric acid
	3-Methylcrotonoylglycine
	3-Sulfinoalanine
	4-Aminobenzoic acid
	4-Aminobutyric acid
	4-Hydroxyproline
	5-Hydroxy-tryptophan
	5-Oxoproline
	Acetylglycine
	Dimethylglycine
	Dopa
	Glutamic acid 5-methylester
	Glycyl-Glycine
	Hexanoylglycine
	Histidinol
	Homocysteine
	Homocystine
	Homoserine
	Hydroxylysine
	Hypotaurine
	Isobutyrylglycine
	Isovalerylglycine
	Kynurenine
	N6-Acetyllysine
	N-Acetylaspartic acid
	N-Acetylglutamine
	N-Acetylglutamine
	N-Acetyl-Lysine
	N-Acetyl-Ornithine
	N-Acetylserine
	N-Acetyltyrosine
	N-Butyrylglycine
	Norvaline
	O-Acetylserine
	O-Phospho-Serine
	Pantothenic acid
	Propionylglycine
	Sarcosine
	S-Benzyl-Cysteine
	Suberylglycine
	Taurine
	Threo-b-hydroxyaspartic acid
	Tiglylglycine

Smart Metabolites Database enables the measurement of 376 hydrophilic metabolic compounds, including the metabolites shown above.
GC-MS(/MS) systems combined with headspace sampling and solid phase micro extraction (SPME) enable the measurement of volatile metabolites.

LC/MS/MS Method Package for Lipid Mediators
Index of Compounds

No.	CAT	Compound
1	LA	(±)12,13-DiHOME
2	LA	(±)9,10-DiHOME
3	LA	13(S)-HODE
4	LA	9(S)-HODE
5	LA	(±)9-HpODE
6	LA	13-OxoODE
7	LA	13(S)-HpODE
8	LA	9-OxoODE
9	LA	(±)12(13)-EpOME
10	LA	(±)9(10)-EpOME
11	ALA	9(S)-HOTrE
12	ALA	13(S)-HOTrE
13	ALA	13(S)-HpOTrE
14	EDA	11(S)-HEDE
15	EDA	(±)15-HEDE
16	EDA	15-OxoEDE
17	AA	tetranor-PGFM
18	AA	tetranor-PGEM
19	AA	tetranor-PGDM
20	AA	tetranor-PGJM
21	AA	tetranor-PGAM
22	AA	20-hydroxy Prostaglandin $F_{2\alpha}$
23	AA	20-hydroxy Prostaglandin E_2
24	AA	18-carboxy dinor LTB_4
25	AA	13,14-dihydro-15-keto-tetranor Prostaglandin $F_{1\beta}$
26	AA	2,3-dinor-8-iso Prostaglandin $F_{2\alpha}$
27	AA	2,3-dinor Thromboxane B_2
28	AA	13,14-dihydro-15-keto-tetranor Prostaglandin $F_{1\alpha}$
29	AA	2,3-dinor-11β-Prostaglandin $F_{2\alpha}$
30	AA	6-keto-Prostaglandin $F_{1\alpha}$
31	AA	13,14-dihydro-15-keto-tetranor Prostaglandin D_2
32	AA	20-carboxy leukotriene B_4
33	AA	20-hydroxy leukotriene B_4
34	AA	11-dehydro-2,3-dinor Thromboxane B_2
35	AA	13,14-dihydro-15-keto-tetranor Prostaglandin E_2
36	AA	6,15-diketo-13,14-dihydro Prostaglandin $F_{1\alpha}$
37	AA	$iPF_{2\alpha}$-IV
38	AA	8-iso-15(R)-Prostaglandin $F_{2\alpha}$
39	AA	8-iso Prostaglandin $F_{2\alpha}$
40	AA	Thromboxane B_2
41	AA	11β-Prostaglandin $F_{2\alpha}$
42	AA	(±)5-$iPF_{2\alpha}$-VI
43	AA	8-iso-15-keto Prostaglandin $F_{2\alpha}$
44	AA	Prostaglandin $F_{2\alpha}$
45	AA	8-iso-13,14-dihydro-15-keto Prostaglandin $F_{2\alpha}$
46	AA	8-iso Prostaglandin E_2
47	AA	Prostaglandin E_2
48	AA	11-dehydro Thromboxane B_2
49	AA	15-keto Prostaglandin $F_{2\alpha}$
50	AA	11β-Prostaglandin E_2
51	AA	5(S),14(R)-LXB_4
52	AA	Prostaglandin K_2
53	AA	Prostaglandin D_2
54	AA	15-keto Prostaglandin $F_{1\alpha}$
55	AA	11β-13,14-dihydro-15-keto Prostaglandin $F_{2\alpha}$
56	AA	15-keto Prostaglandin E_2
57	AA	13,14-dihydro Prostaglandin $F_{1\alpha}$
58	AA	14,15-LTC_4
59	AA	13,14-dihydro-15-keto Prostaglandin $F_{2\alpha}$
60	AA	5(S),6(R)-Lipoxin A_4
61	AA	13,14-dihydro-15-keto Prostaglandin E_2
62	AA	5(S),6(S)-Lipoxin A_4
63	AA	14,15-LTE4, Eoxin E_4
64	AA	13,14-dihydro-15-keto Prostaglandin D_2
65	AA	Leukotriene C_4
66	AA	11-trans LTC_4
67	AA	Leukotriene D_4
68	AA	Leukotriene E_4
69	AA	Leukotriene F_4
70	AA	8-iso Prostaglandin A_2
71	AA	11-trans LTD_4
72	AA	Prostaglandin A_2
73	AA	Prostaglandin J_2
74	AA	11-trans LTE_4
75	AA	Prostaglandin B_2
76	AA	8,12-iso-$iPF_{2\alpha}$-VI 1,5- lactone
77	AA	8(S),15(S)-DiHETE
78	AA	6-trans LTB_4
79	AA	5(S),15(S)-DiHETE
80	AA	13,14-dihydro-15-keto Prostaglandin A_2
81	AA	Leukotriene B_4
82	AA	13,14-dihydro-15-keto Prostaglandin J_2
83	AA	12-oxo LTB_4
84	AA	tetranor-12(S)-HETE
85	AA	N-acetyl LTE_4
86	AA	Leukotriene B_3
87	AA	(±)14(15)-DiHET
88	AA	12(S)-HHTrE
89	AA	(±)11(12)-DiHET
90	AA	(±)8(9)-DiHET
91	AA	20-carboxy arachidonic acid
92	AA	(±)5(6)-DiHET
93	AA	19(S)-HETE
94	AA	15-deoxy-delta12,14-PGJ_2
95	AA	20-HETE
96	AA	(±)18-HETE
97	AA	(±)17-HETE
98	AA	(±)16-HETE
99	AA	15(S)-HETE
100	AA	11(S)-HETE
101	AA	8(S)-HETE
102	AA	15-OxoETE
103	AA	15(S)-HpETE
104	AA	12(S)-HETE
105	AA	(±)9-HETE
106	AA	5(S)-HETE
107	AA	12(S)-HpETE
108	AA	12-OxoETE
109	AA	(±)5,6-DHET-lactone
110	AA	5(S)-HpETE
111	AA	(±)14(15)-EET
112	AA	5-OxoETE
113	AA	(±)11(12)-EET
114	AA	(±)8(9)-EET
115	AA	(±)5(6)-EET
116	AA	Arachidonic Acid (AA)
117	ADA	1a,1b-dihomo-Prostaglandin $F_{2\alpha}$
118	DGLA	2,3-dinor Thromboxane B_1
119	DGLA	6-keto Prostaglandin E_1
120	DGLA	2,3-dinor Prostaglandin E_1
121	DGLA	Thromboxane B_1
122	DGLA	8-iso Prostaglandin $F_{1\alpha}$
123	DGLA	Prostaglandin $F_{1\alpha}$
124	DGLA	8-iso Prostaglandin E_1
125	DGLA	Prostaglandin E_1
126	DGLA	Prostaglandin D_1
127	DGLA	13,14-dihydro Prostaglandin E_1
128	DGLA	13,14-dihydro-15-keto Prostaglandin D_1
129	DGLA	8-iso Prostaglandin A_1
130	DGLA	Prostaglandin A_1
131	DGLA	15(S)-HETrE
132	DGLA	8(S)-HETrE
133	DGLA	5(S)-HETrE
134	EPA	Δ17-6-keto Prostaglandin $F_{1\alpha}$
135	EPA	Resolvin E_1
136	EPA	8-iso Prostaglandin $F_{3\alpha}$
137	EPA	Thromboxane B_3
138	EPA	Prostaglandin $F_{3\alpha}$
139	EPA	11-dehydro Thromboxane B_3
140	EPA	Prostaglandin E_3
141	EPA	Prostaglandin D_3
142	EPA	Lipoxin A_5
143	EPA	Leukotriene B_5
144	EPA	(±)17,18-DiHETE
145	EPA	(±)14(15)-DiHETE
146	EPA	(±)5(6)-DiHETE
147	EPA	(±)18-HEPE
148	EPA	15(S)-HEPE
149	EPA	11(S)-HEPE
150	EPA	8(S)-HEPE
151	EPA	9(S)-HEPE
152	EPA	12(S)-HEPE
153	EPA	5(S)-HEPE
154	EPA	15(S)-HpEPE
155	EPA	12(S)-HpEPE
156	EPA	5(S)-HpEPE
157	EPA	(±)17(18)-EpETE
158	EPA	(±)14(15)-EpETE
159	EPA	Eicosapentaenoic Acid(EPA)
160	DHA	Resolvin D_3
161	DHA	Resolvin D_2
162	DHA	Resolvin D_1
163	DHA	Resolvin D_4
164	DHA	7(R)-Maresin 1
165	DHA	10(S),17(S)-DiHDHA
166	DHA	Resolvin D_5
167	DHA	7(S),17(S)-hydroxy-docosapentaenoic acid
168	DHA	(±)19(20)-DiHDPA
169	DHA	(±)20-HDHA
170	DHA	(±)16-HDHA
171	DHA	(±)17-HDHA
172	DHA	(±)13-HDHA
173	DHA	(±)10-HDHA
174	DHA	(±)14-HDHA
175	DHA	(±)11-HDHA
176	DHA	(±)7-HDHA
177	DHA	(±)8-HDHA
178	DHA	17(S)-HpDHA
179	DHA	(±)4-HDHA
180	DHA	(±)19(20)-EpDPA
181	DHA	(±)16(17)-EpDPA
182	DHA	Docosahexaenoic Acid (DHA)
183	EA	Prostagrandin $F_{2\alpha}$ Ethanolamide
184	EA	Prostagrandin E_2 Ethanolamide
185	EA	Prostagrandin E_1 ethanolamide
186	EA	Prostagrandin D_2 Ethanolamide
187	EA	LTB_4 ethanolamide
188	EA	(±)14(15)-EET ethanolamide
189	EA	(±)11(12)-EET ethanolamide
190	EA	(±)8(9)-EET ethanolamide
191	EA	(±)5(6)-EET ethanolamide
192	EA	Arachidonoyl ethanolamide
193	EA	OEA (oleoyl ethanolamide)
194		Lyso-PAF C-16
195		PAF C-16
196		Azelaoyl PAF
197	ISTD	tetranor-PGEM-d_6
198	ISTD	6-keto-Prostaglandin $F_{1\alpha}$-d_4
199	ISTD	Thromboxane B_2-d_4
200	ISTD	Prostaglandin $F_{2\alpha}$-d_4
201	ISTD	Prostaglandin E_2-d_4
202	ISTD	Prostaglandin D_2-d_4
203	ISTD	Leukotriene C_4-d_5
204	ISTD	Leukotriene D_4-d_5
205	ISTD	Prostaglandin A_2-d_4
206	ISTD	Leukotriene B_4-d_4
207	ISTD	(±)14(15)-DiHET-d_{11}
208	ISTD	5(S) HETE-d_8
209	ISTD	12(S)-HETE-d_8
210	ISTD	5(S)-HETE-d_8
211	ISTD	PAF C-16-d_4
212	ISTD	(±)11(12)-EET-d_{11}
213	ISTD	Oleoyl ethanolamide-d_4
214	ISTD	AA-d_8

Category codes

LA : linoleic acid
ALA : α-linolenic acid
EDA : eicosadienoic acid
AA : arachidonic acid
ADA : adrenic acid
DGLA : dihomo-γ-linolenic acid
EPA : eicosapentaenoic acid
DHA : docosahexaenoic acid
EA : ethanolamide
ISTD : internal standard

LC/MS/MS MRM Library for Metabolic Enzymes in Yeast
Index of Compounds

AAT1	ADH4	ARO4	CYS3	GAL10	GPM1	HXK2	LPD1	MET22	PGM1	SAM2	THR4
AAT2	ADH6	ARO7	CYS4	GAL7	GPM2	ICL1	LSC1	MET3	PGM2	SAM4	TKL1
ACH1	ADK1	ARO8	DAK1	GCV1	GPM3	IDH1	LSC2	MET6	PRO1	SDH1	TKL2
ACO1	AGX1	ASN1	DAL7	GCV2	GSY1	IDH2	LYS1	MHT1	PRO2	SDH2	TPI1
ACO2	ALD3	ASN2	DUR1,2	GCY1	GSY2	IDP1	LYS12	MLS1	PRO3	SDH3	TPS1
ACS1	ALD4	ASP1	ECM17	GDB1	GUA1	IDP2	LYS2	MVD1	PRS1	SDH4	TPS2
ACS2	ALD5	ATH1	ECM40	GDH1	GUK1	IDP3	LYS20	NQM1	PRS2	SER1	TPS3
ADE1	ALD6	BAT1	ENO1	GDH2	GUT2	ILV1	LYS21	NTH1	PRS3	SER2	TRP1
ADE12	ALT1	BAT2	ENO2	GDH3	HIS1	ILV2	LYS4	PCK1	PRS4	SER3	TRP2
ADE13	ALT2	BNA3	ERG10	GLC3	HIS3	ILV3	LYS9	PDA1	PRS5	SER33	TRP3
ADE16	ARG1	BNA5	ERG13	GLK1	HIS4	ILV5	MAE1	PDB1	PYC1	SFA1	TRP5
ADE17	ARG2	CAR1	ERG20	GLN1	HIS5	IMD2	MDH1	PDC1	PYC2	SHM2	TSL1
ADE2	ARG3	CAR2	ERR	GLT1	HIS6	IMD4	MDH2	PDC5	RHR2	SOL3	UGA1
ADE4	ARG4	CDC19	FBA1	GLY1	HIS7	KGD1	MDH3	PDC6	RKI1	SOL4	UGA2
ADE5,7	ARG5,6	CIT1	FBP1	GND1	HOM2	KGD2	MET10	PDE1	RNR2	TAL1	UGP1
ADE6	ARG8	CIT2	FRD1	GND2	HOM3	LAT1	MET14	PFK1	RNR4	TDH1	URA2
ADH1	ARO1	CIT3	FUM1	GPD1	HOM6	LEU1	MET16	PFK2	RPE1	TDH2	YNK1
ADH2	ARO2	CPA1	GAD1	GPD2	HOR2	LEU2	MET17	PGI1	SAH1	TDH3	YPR1
ADH3	ARO3	CPA2	GAL1	GPH1	HXK1	LEU4	MET2	PGK1	SAM1	THR1	ZWF1

LC/MS/MS Method Package for Bile Acids
Index of Compounds

Abbreviation	Common name	Class	Abbreviation	Common name	Class
• αMCA	a-Muricholic acid	PBA	• GDCA	Glycodeoxycholic acid	G SBA
• βMCA	b-Muricholic acid	PBA	• GHDCA	Glycohyodeoxycholic acid	G SBA
• CA	Cholic acid	PBA	• GLCA	Glycolithocholic acid	G SBA
• ωMCA	w-Muricholic acid	PBA	• GUDCA	Glycoursodeoxycholic acid	G SBA
• 12-keto-LCA	12-Keto-deoxycholic acid	SBA	• TCDCA	Taurochenodeoxycholic acid	T SBA
• 7-keto-DCA	7-Keto-deoxycholic acid	SBA	• TDCA	Taurodeoxycholic acid	T SBA
• 7-keto-LCA	7-Keto-lithocholic acid	SBA	• THDCA	Taurohyodeoxycholic acid	T SBA
• allo-CDCA	Allo-chenodeoxycholic acid	SBA	• TLCA	Taurolithocholic acid	T SBA
• allo-LCA	Allo-lithocholic acid	SBA	• TUDCA	Tauroursodeoxycholic acid	T SBA
• CDCA	Chenodeoxycholic acid	SBA	• D4-CDCA	Chenodeoxycholic acid-2,2,4,4-d4	ISTD
• DCA	Deoxycholic acid	SBA	• D4-CA	Cholic acid-2,2,4,4-d4	ISTD
• HDCA	Hyodeoxycholic acid	SBA	• D4-DCA	Deoxycholic acid-2,2,4,4-d4	ISTD
• LCA	Lithocholic acid	SBA	• D4-GCA	Glycocholic acid-2,2,4,4 -d4	ISTD
• UDCA	Ursodeoxycholic acid	SBA	• D4-GDCA	Glycodeoxycholic acid-2,2,4,4 -d4	ISTD
• GCA	Glycocholic acid	G PBA	• D4-GLCA	Glycolithocholic acid-2,2,4,4-d4	ISTD
• TαMCA	Tauro-a-muricholic acid	T PBA	• D4-LCA	Lithocholic acid-2,2,4,4-d4	ISTD
• TβMCA	Tauro-b-muricholic acid	T PBA	• D4-TCDCA	Taurochenodeoxycholic acid-2,2,4,4 -d4	ISTD
• TCA	Taurocholic acid	T PBA	• D5-TCA	Taurocholic acid-2,2,3,4,4-d5	ISTD
• GCDCA	Glycochenodeoxycholic acid	G SBA	• D5-TLCA	Taurolithocholic acid-2,2,3,4,4-d5	ISTD

Class codes

PBA: Primary bile acid
SBA: Secondary bile acid
T PBA: Taurine-conjugated primary bile acid
T SBA: Taurine-conjugated secondary bile acid
G PBA: Glycine-conjugated primary bile acid
G SBA: Glycine-conjugated secondary bile acid
ISTD: Internal standard

List of Main Applications

No.	Application Title	Analysis Target	Sample	Instrument
M274	Construction of a Regression Model for a Coffee Sensory Evaluation Through the Comprehensive Analysis of Metabolites	Metabolite	Food	GCMS
M271	Investigating Food Quality Evaluation: Complete Analysis of Aroma Compounds and Metabolites in Food	Metabolite	Food	GCMS
Application Note No.48	Comprehensive Measurement of Metabolites Using GC-MS/MS and LC-MS/MS —An Application to the Research of the Intestinal Environment—	Metabolite	Fecal	GCMS, LCMS
C157	A Multiomics Approach Using Metabolomics and Lipidomics	Metabolite, phospholipid	Microbial culture medium	LCMS
C134	Multi-Component Analysis of Five Beers	Metabolite	Food	LCMS
C132	Comprehensive Analysis of Primary and Secondary Metabolites in Citrus Fruits Using an Automated Method Changeover UHPLC System and LC/MS/MS System	Metabolite	Food	LCMS
C131	Application of Metabolomics to Microbial Breeding	Metabolite	Microbial culture medium	LCMS
C186	Comprehensive Cell Culture Profiling Using the LCMS-9030 Quadrupole TOF Mass Spectrometer	Metabolite	Cell culture medium	LCMS
C106	Simultaneous Analysis of Culture Supernatant of Mammalian Cells Using Triple Quadrupole LC/MS/MS	Metabolite	Cell culture medium	LCMS
C192	Simultaneous Analysis of Chiral Amino Acids Produced by Intestinal Bacteria by LC/MS/MS	Chiral amino acid	Fecal, blood	LCMS
C156	Analysis of Chiral Amino Acids within Fermented Beverages Utilizing a Column Switching System	Chiral amino acid	Food	LCMS
C149	Developing a Chiral Amino Acid Analysis Method That Uses Column Switching	Chiral amino acid	Reference material	LCMS
C168	Analysis of Short-Chain Fatty Acids/Organic Acids (3-NPH Derivatives) in Fecal Specimens from SPF and Antibiotic-Fed Mice	Short chain fatty acid	Fecal	LCMS
C182	A Method of Simultaneous Analysis for 196 Lipid Mediators and Related Compounds Using Triple Quadrupole LC/MS/MS	Lipid mediator	Blood	LCMS
C113	Lipid Mediator Profiling of Human Serum Using the Triple Quadrupole LC/MS/MS	Lipid mediator	Blood	LCMS
C155	Phospholipid Analysis for Four Types of Mouse Tissues	Lipid mediator	Tissue	LCMS
C151	Phospholipid Analysis Using SimLipid Software	Phospholipid	Tissue	LCMS
C137	Development of a Phospholipid Profiling Method Using Triple Quadrupole LC/MS/MS	Phospholipid	Tissue	LCMS
B88	Establishment of a Method for Direct Analysis of the Mouse Liver Metabolome Using the DPiMS-8060	Metabolite	Tissue	DPiMS
B87	Establishment of a Method for Direct Analysis of the Mouse Brain Metabolome Using the DPiMS-8060	Metabolite	Tissue	DPiMS

*For the latest applications, please contact the Shimadzu sales representative in your country/region or visit our website.

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