# Ionization-assisting substrates

Desorption Ionization Using Through Hole Alumina MEmbrane









# DIUTHAME ensures high repro ducibility and accuracy in your mass spectrometry tasks!



Hamamatsu offers ionization-assisting substrates called DIUTHAME that support ionization in mass spectrometry in place of matrix that is currently used for MALDI (Matrix-Assisted Laser Desorption/Ionization) and also eliminate the cumbersome sample pretreatment process. DIUTHAME brings high reproducibility and ease-of-handling to mass spectrometry by serving as a completely new tool that can readily be used by all MALDI mass spectrometer users.

# •Example: Difference in preparations for frozen section measurements of mouse brain





End of measurement



Ionization-assisting substrates DIUTHAME

# Mass spectrum measurement example

#### PEO-hydrogenated castor oil

Measurement sample: Cosmetic raw material (Hydrogenated castor oil)

#### Measurement example of PEO-hydrogenated castor oil using DIUTHAME





#### Measurement method

The mixed sample was dropped from above the DIUTHAME.

#### PEO-monostearate

Measurement sample: Industrial surfactant (PEO monostearate)



#### Measurement method

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The mixed sample was dropped from above the DIUTHAME.

# NO matrix required

- NO matrix background noise
- High reproducibility with minimal variation no matter who does the measurement
- NO sample pretreatment required
- High spatial resolution in imaging mass spectrometry ensured by nanometer-order structure

# **How DIUTHAME** differs from conventional MALDI

**Features** 

Item	DIUTHAME	MALDI
Background noise	None	Matrix noise appears
Ease of handling	Easy	Expertise is required
Reproducibility	High	Not so high
Spatial resolution	High	Not so high
Measurement of high molecules	Possible depending on samples	Possible





#### Measurement sample details

The sample was dissolved in THF at a concentration of 1 mg/mL. NaTFA was used as the cationizing agent and dissolved in THF at a concentration of 1 mg/mL.

The sample was mixed with the cationizing agent at a ratio of 1:10 (v/v).

m/z

# Mass spectrum measurement example

### Angiotensin II

Measurement sample: Angiotensin II ([M+H]<sup>+</sup>, m/z 1046.5): 1 µM Measurement conditions: Linear, positive ion mode



#### Measurement method

The mixed sample was soaked up from below the DIUTHAME.

#### Measurement sample details

Angiotensin II: DHC : CitAc : ACN=1 : 1 : 1 : 1 Angiotensin II: 1 µM DHC (Diammonium hydrogen citrate): 0.2 M CitAc (Citric acid): 0.2 M ACN: Acetonitrile

#### Polyethylene glycol 2000

Measurement sample: Polyethylene glycol 2000: 1mM in acetone Measurement conditions: Linear, positive ion mode



#### Measurement method

The mixed sample was dropped from above the DIUTHAME.

#### Insulin

Measurement sample: Insulin ([M+H]+, m/z 5733.6): 0.5 mM Measurement conditions: Linear, positive ion mode



#### **Measurement method**

The mixed sample was soaked up from below the DIUTHAME.

#### Measurement sample details

Insulin: DHC : CitAc : ACN=1 : 1 : 1 : 1 Insulin: 0.5 mM DHC (Diammonium hydrogen citrate): 0.2 M CitAc (Citric acid): 0.2 M ACN: Acetonitrile

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# Measurement examples using imaging mass spectrometry

<Microscopic image>

\* After making measurements using imaging mass spectrometry, a microscopic image was captured from above the DIUTHAME by using a microscope.

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#### Black rice



m/z 920 (Phosphatidylcholine) Measurement conditions: Linear, positive ion mode Laser pitch: 50 µm

#### Measurement method

- ①Set a slice of black rice on an ITO glass slide.
- 2Place the DIUTHAME substrate on the sliced black rice.
- 3 Drop 2 µL of "70 % AcCN / 30 % H20" solution from above the DIUTHAME to extract the components of interest. (4) After the sample dries,
- make measurements using imaging mass spectrometry



#### Mouse brain



*m/z* 840 Measurement conditions: Linear, positive ion mode Laser pitch: 60 µm

(1)

**Measurement method** 

①Set a slice of frozen mouse brain on an ITO glass slide.

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3 After the brain slice thaws, the components derived from the sample are soaked up by capillary action.

(2)

2 Place the DIUTHAME on the mouse brain slice before it thaws.

(4) After the sample dries, make measurements using imaging mass spectrometry.



\* Before making measurements using imaging mass spectrometry, a microscopic image was captured from above the DIUTHAME by using a microscope

3 The components of interest are soaked up 4

/+ + + +

glass



*m/z* 910 Measurement conditions: Linear, negative ion mode Laser pitch: 60 µm



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First time

## • Comparison between MALDI and DIUTHAME using mouse brain (m/z 848)

#### MALDI



Tissue section has contracted somewhat

10 um

50 µm

# Cocoa raw bean (Triglycerides)

Measurement sample: Triglycerides in cocoa raw beans Measurement conditions: Linear, positive ion mode



#### Measurement method

①Set a slice of cacao raw bean on an ITO glass slide. 2Place the DIUTHAME on the sliced bean. 3Drop 2 µL of acetone from above the DIUTHAME to extract the components of interest.

④Irradiate a laser beam onto the DIUTHAME to cause ionization.



Olmaging mass spectrometry measurement example

Measurements were carried out in cooperation with Designated Assistant Professor Keiko Kuwata, The Institute of Transformative Bio-Melecules Nagoya University

### • Reproducibility of serial slices of mouse brain (near *m/z* 850)



Mouse brain slice thickness: 50 µm Laser pitch: 50 µm



Second time



#### Third time

<Conditions> Mouse brain slice thickness

Matrix conditions DHB 50 mg/ml, 50 % ACN Spray coating Measurement conditions Reflectron, positive ion mode Laser pitch

## DIUTHAME



Good spatial resolution 50 µm Maintains shape of tissue section

<Conditions> Mouse brain slice thickness 50 um DIUTHAME size Effective diameter: 18 mm Measurement conditions Reflectron positive ion mode Laser pitch

### A13331-3-1 (3 mm diameter) For mass spectrum



## A14111-3-1 (3 mm diameter × 9 ch) For mass spectrum





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