

# Development of blotting method using DIUTHAME for imaging MS

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## 1. Introduction

We have developed DIUTHAME (Desorption Ionization Using Through Hole Alumina MEmbrane) as an alternative to matrices. Ionization process using DIUTHAME and a comparison of the features with MALDI are shown in Fig.1 and Table 1.

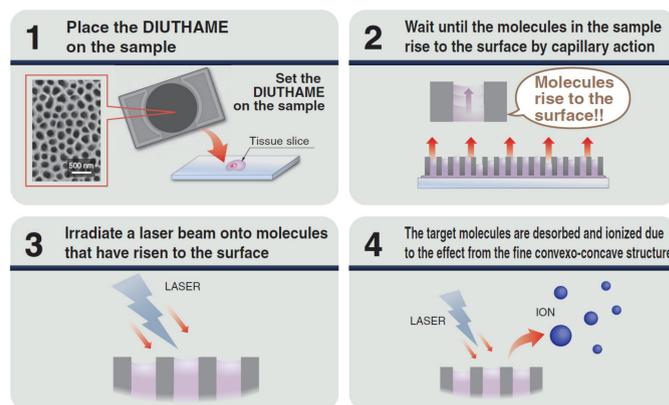


Fig.1: Ionization process using DIUTHAME

Table 1: a comparison of the features with MALDI

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
Ionization of large molecules	Possible depending on samples	Possible

Table 2 : Strawberry shipping standard

Standard	Single grain weight [g]
DX	27
3L	25
2L	15
L	11
M	9
S	7

Difficult to thin sectioning

About 30g

## 3-1. Results: positive/negative mode

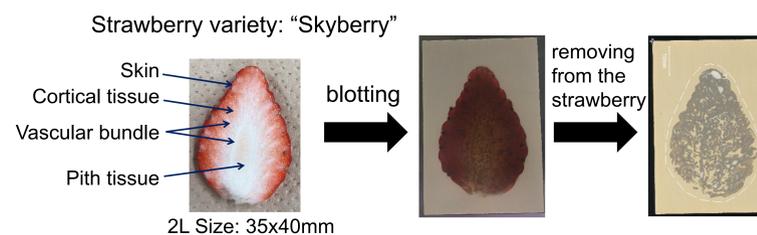
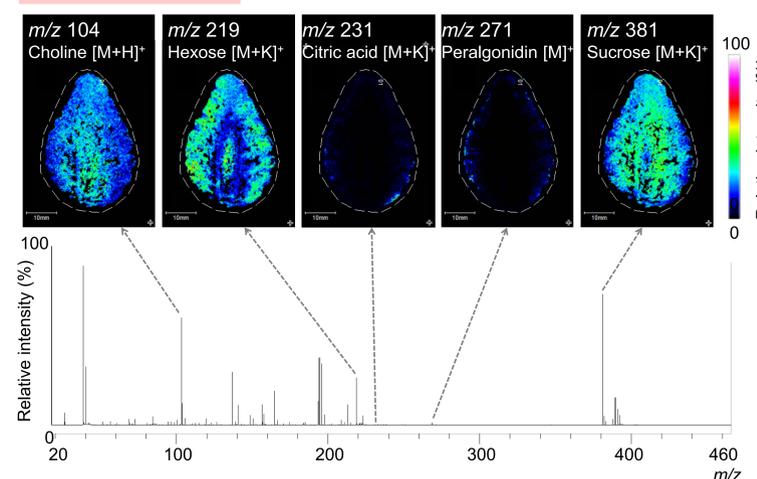


Fig.3: Optical image in each step

From the results of imaging MS of blotting onto DIUTHAME chip, the distribution of:

- choline was around pith tissue.
- hexose was mainly observed in cortical tissue and pith tissue in the positive mode and observed all over the strawberry section in the negative mode.
- citric acid was observed in the skin in the positive mode and observed in the tip side and cortical tissue in the negative mode.
- pelargonidin and  $m/z$  301 attributable to quercetin and/or ellagic acid were located in the skin of a strawberry.
- sucrose was predominantly observed in the tip side of cortical tissue and vascular bundle.
- malic acid was similar to that of citric acid in the negative mode. This characteristic pattern was presumably formed by the reticulated juice after drying.

### Positive mode



### Negative mode

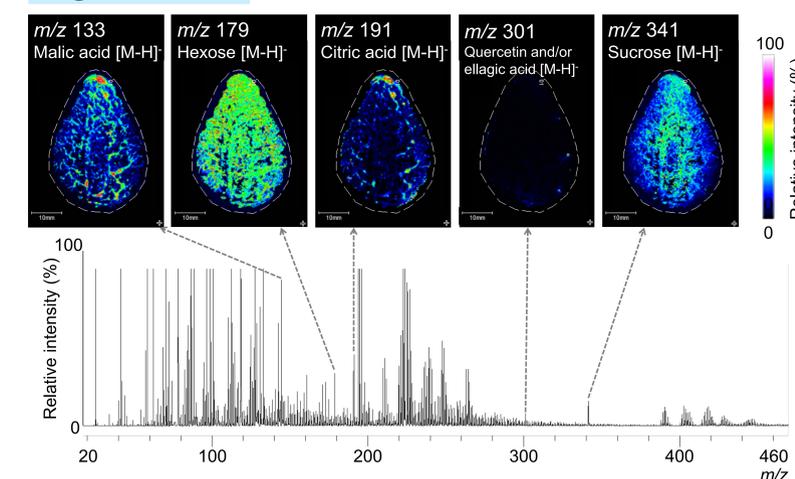


Fig.4: Imaging analysis of strawberry using blotting onto DIUTHAME chip

## 2. Methods

To conduct imaging MS, a raw strawberry was cut in half with knife under the room temperature. Then DIUTHAME chip was put on the cut surface and the contained materials on the strawberry were blotted. The DIUTHAME chip that was carefully removed from the strawberry, was then directly attached on MTP TLC Adapter (BigSlides for MALDI, Bruker). Imaging MS was taken by ultrafleXtreme (Bruker) MALDI time-of-flight mass spectrometer. Ion maps of various molecules were created by flexImaging software (Bruker).

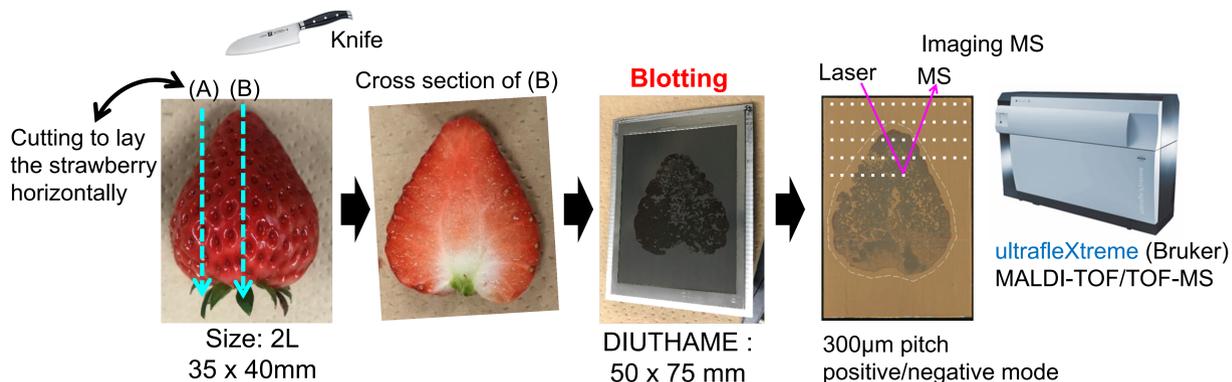


Fig.2: Workflow of blotting onto DIUTHAME chip in imaging MS

## 3-2. Results: effect of cutting direction

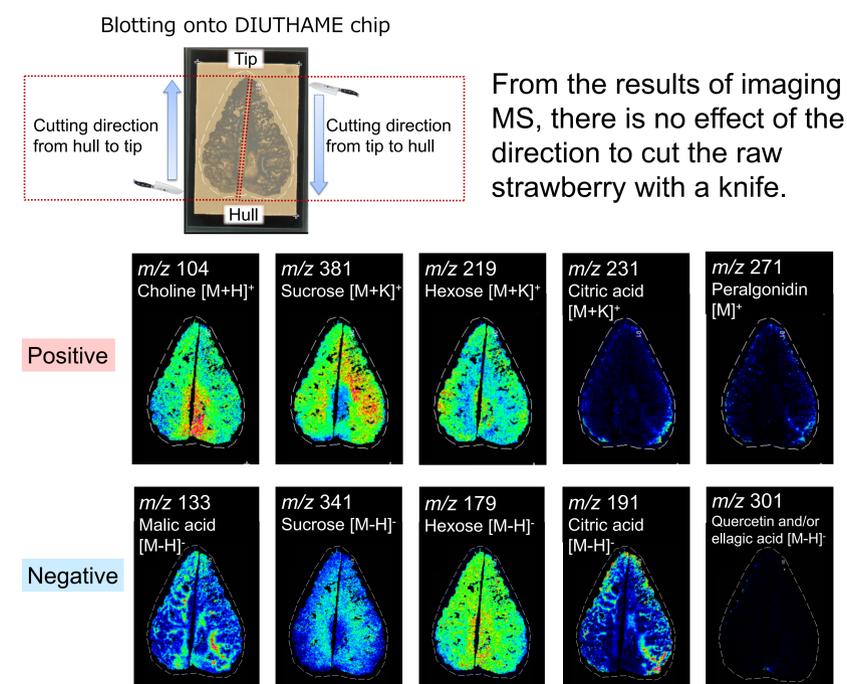


Fig.5: Imaging results obtained by different cutting directions

## 4. Conclusion

- Imaging MS results of blotting onto DIUTHAME chip were consistent with MALDI imaging MS using frozen section.
- We suggest that **the blotting method using DIUTHAME chip is very easy and effective** for imaging MS of those which thin sectioning is difficult for.

### References

Enomoto et al., *J Agric Food Chem*, 66, 4958-4965, 2018.



For inquiries about DIUTHAME, Visit at **Booth #110**