# Identification of peach species using the X500R QTOF system

Gu Ye<sup>1</sup>, Mu Pengqian<sup>2</sup>, Yang Zong<sup>2</sup>, Liu Bingjie<sup>2</sup>, Guo Lihai<sup>2</sup> <sup>1</sup>Wuxi Food Safety Inspection and Test Center, Wuxi, China ; <sup>2</sup>SCIEX Asia Pacific Application Support Center, Shanghai, China

#### INTRODUCTION

Peaches are an important stone fruit crop grown all over the world, and they are both nutritionally and economically important. Peaches are rich in fructose, dietary fiber, polyphenols, trace elements and other nutrients. Understanding the differences in metabolites between cultivars is critical for nutritional studies of peaches. Accurate mass spectrometry systems have proven valuable for qualitative analysis. In this study, we collected four kinds of peaches: honey peach, flat peach, nectarine and yellow peach, to evaluate their metabolite differences by liquid chromatography-high resolution mass spectrometry.

### MATERIALS AND METHODS

**Sample preparation:** Homogenized peaches were placed in a centrifuge tube, acetonitrile was added for extraction, then anhydrous sodium sulfate was added. After mixing, the acetonitrile layer was removed and the sample was dried, then resuspended in 50% methanol in water. The sample was injected onto the SCIEX X500R QTOF system coupled with an ExionLC system.

HPLC conditions: A 20-minute gradient on Phenomenex Kinetex C18 column (2.6  $\mu$ m, 2.1 × 100 mm) was employed to ensure high sensitivity and good separation. Composition of mobile phases was 2 mM ammonium acetate in water/methanol. The flow rate of 300  $\mu$ L/min. The injection volume was set to 5  $\mu$ L.

MS/MS conditions: TOF MS data and MS/MS data were acquired in one injection using the information-dependent acquisition (IDA) scan mode with dynamic background subtraction (DBS) enabled. The MS source conditions were optimized as follows: curtain gas (CUR), 30 psi; collision gas (CAD), medium; nebulizing gas (GS1), 55 psi; heater gas (GS2), 55 psi; ion spray voltage (IS), 5500V/-4500V in positive mode /negative mode; and source temperature, 550°C. Other mass spectrometry parameters are in Table 1. Data processing was performed using SCIEX OS software 2.1, and principal component analysis (PCA) in MarkerView software.

# Mass

**RESULTS** The high-speed and high-sensitivity IDA-DBS data acquisition approach, in both

positive and negative mode, effectively triggered MS/MS of compounds of interest in peaches. TOF MS data and MS/MS data were acquired in one injection (for each polarity). Using the SCIEX OS software and the high-resolution natural products and metabolite libraries, the main components of peaches were quickly identified, and the target could be confirmed automatically according to mass error, isotope distribution, and MS/MS spectrum, to ensure the accuracy and reliability of identification results. The identification results showed that the mass deviation of most compounds was within 1 ppm (Figure 1). The matching of isotopes and MS/MS spectra was high, showing the excellent stability and accuracy of the X500R QTOF system. Some 81 compounds in peaches, including phenols, vitamins and organic acids were quickly identified from the positive and negative ion data.

#### Table 1. Mass spectrometry parameters.

pectrometry parameters	Settings
TOF MS (Da)	100 - 1000
TOF MS/MS (Da)	50 - 1000
Dynamic Background Substraction	On
Product of Candidate MS/MS per cycle	12



Figure 1. Partial screening results of target compounds by SCIEX OS software.

Using MarkerView software, PCA analysis and t-test analysis were performed on the identified 81 compounds in peaches to find differential compounds. The PCA analysis results showed that the 4 different kinds of peaches could be effectively distinguished (Figure 2). Through t-test analysis, 12 compounds in honey peach were significantly different from those of other species (Figure 3). Honey peach is rich in organic acids, chlorogenic acid, catechins, and proanthocyanidins. In addition, 16 different markers were found in flat peach, nectarine and yellow peach – mainly polyphenols and organic acids.



Figure 2. Statistical analysis of the four kinds of peaches using MarkerView software. Principal Component Analysis (PCA) was performed using the quantitative results from the identified compounds. The scores plot on the left shows that in both cases the samples from the different types of peaches clearly separate, with replicates clustering tightly together. Using the Loadings plot on the right, the compounds responsible for the separation can be determined.





**Figure 3. T-test.** A t-test can also be performed which provides an additional way to determine specific compounds that differ significantly between the different peaches. Shown is chlorogenic acid, significant higher in the honey peach samples.

# **CONCLUSIONS**

A high efficiency and sensitive method for evaluation of metabolite differences in four kinds of peaches was successfully achieved.

# **TRADEMARKS/LICENSING**

The SCIEX clinical diagnostic portfolio is For In Vitro Diagnostic Use. Rx Only. Product(s) not available in all countries. For information on availability, please contact your local sales representative or refer to www.sciex.com/diagnostics. All other products are For Research Use Only. Not for use in Diagnostic Procedures.

www.sciex.com/trademarks). © 2022 DH Tech. Dev. Pte. Ltd. RUO-MKT-10-14738-A

•	n1 = 6	n2 = 18										
oup	Use	t-value	p-value /	Mean 1	Mean 2	Median 1	Median 2	Sigma 1	Sigma 2	Delta	Fold Change	Log (F
		136.48	1.0364e-33	1.092e7	5.787e4	1.095e7	2.440e4	3.312e5	6.790e4	1.086e7	1.886e2	2.276e
	$\checkmark$	89.61	1.0673e-29	1.430e5	1.829e3	1.430e5	0.000e0	4.899e3	2.719e3	1.412e5	7.817e1	1.893e
	$\checkmark$	88.10	1.5471e-29	2.782e6	9.988e4	2.745e6	8.720e4	9.152e4	5.415e4	2.682e6	2.785e1	1.445e
	$\checkmark$	74.65	5.8504e-28	1.742e4	0.000e0	1.725e4	0.000e0	1.038e3	0.000e0	1.742e4		
	$\checkmark$	68.50	3.8518e-27	1.257e6	1.204e4	1.260e6	7.805e3	7.967e4	7.473e3	1.245e6	1.044e2	2.019e
	$\checkmark$	46.39	1.9269e-23	9.283e6	3.896e5	9.310e6	1.490e5	2.330e5	4.451e5	8.894e6	2.383e1	1.377e
		45.43	3.0362e-23	1.895e5	2.690e4	1.885e5	2.445e4	6.950e3	7.771e3	1.626e5	7.045e0	8.479e
	$\checkmark$	36.71	3.1122e-21	5.255e5	6.879e4	5.280e5	5.535e4	1.688e4	2.859e4	4.567e5	7.639e0	8.831e
	$\checkmark$	-30.32	1.9274e-19	4.058e5	9.284e5	4.065e5	9.385e5	8.704e3	4.132e4	-5.226e5	4.371e-1	-3.594
		21 21	3 8851e-16	4 643e4	5 576e3	4 730e4	2 810e3	2 238e3	4 487e3	4 086e4	8.328e0	9 205e 🎽
			_									
			4 <u></u>	岱 掌 🐻	前 🔍 🚍							d <del>an</del>
	YS Kara Kara Kara Kara Kara Kara Kara Kar					)						
	YS 🛉 YS T YS T 1.1			1.1e7 -	t YS YS						Flat peach	
			YS YS	1.0e7 -			YS YS YS				Nec	tarine
				9.0e6 -							Yell	ow peach
				8.0e6 -								
				w 7.0e6 -								
				5 6.0e6 -								
				8 5.0e6 -								
				4.0e6 -								
				3.0e6 -								
рт	YT			2.0e6 -								
Ц_т.	TI YI	Υ		1.0e6 -	F	т		YI		HT		
PT YT	Ý	YT YS	YS	0.0e0 -	Flat	peach	Honey peach	Necta	rine \	fellow peach		
e (by index	c)					-		Group				

Trademarks and/or registered trademarks mentioned herein, including associated logos, are the property of AB Sciex Pte. Ltd. or their respective owners in the United States and/or certain other countries (see

To receive a copy of this poster:

- Scan the code with your phone camera

Complete the form