Enhancing the data quality by high-speed analysis on a single quadrupole mass spectrometer

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1. Introduction

Recently, ultra-high-performance liquid chromatography (UHPLC) has become a popular technique for HPLC and is increasingly used as front separation for mass spectrometry. The benefit of UHPLC was a significant reduction in analytical runtimes with compounds eluting within much narrower peak widths, which in turn necessitated the mass spectrometer to operate faster without compromising data quality. To this end, Shimadzu has developed technologies to enable ultrafast scanning and ultrafast positive/negative switching. In this study, we investigated the impact of the scan speed and the switching speed on the mass chromatogram of some pharmaceutical products using Shimadzu Nexera series and modified LCMS-2020.

Figure 1. the scheme of cycle of data acquisition time and the switching time

2. Material and Reagents

2-1. Instruments

Ultrafast analysis was performed using Nexera XR system consisting of LC-40B XR pumps, SIL-40C XR autosampler, CTO-40C column oven, and SPD-M40 photodiode array detector and modified LCMS-2020 mass spectrometer (Shimadzu Corporation, Kyoto, Japan).

2-2. Materials and Methods

Samples

Pharmaceutical standards (papaverine, propranolol, diphenhydramine, dipyridamole, amitriptyline, reserpine, chloramphenicol, sulfadimethoxine, furosemide, carbamazepine, isopropyl antipyrine, nitrendipine) were purchased from FUJIFILM Wako Pure Chemical Corporation (Japan).

Sample Preparation

Stock solutions of each pharmaceutical (1,000 mg/L) were prepared by dissolving in acetonitrile. The standard mixture solution of pharmaceuticals (10 mg/L) was prepared by mixing all stock solutions and further diluting it with acetonitrile.

Condition of LC and MS

The experimental setting of LC and MS are summarized in Table 1 and Table 2. The MS spectrum with in-source collision-induced dissociation (CID) was acquired with a Qarray voltage of 120 V, much elevated from the default setting.

Table 1. LC conditions

System	: Nexera™ XR
Column	: Shim-pack™ XR-ODS (50 mm x 2.0 mm I.D., 2.2 µm)
Mobile phases	: A: 10 mM ammonium acetate aq. B: Acetonitrile
Flow rate	: 0.3 mL/min
Gradient	: B.Conc 10 % (0-2 min) →30 % (4-6 min) →100 % (20 min)
Colum temperature	: 40 °C
Injection volume	: 1 µL
Detection	: PDA 190-800 nm

Table 2. MS parameters

Ionization	: ESI/APCI (positive and negative mode)
Mode	: SCAN (*m/z*100-1000)
Nebulizing gas flow	: 2.0 L/min
Drying gas flow	: 5.0 L/min
Heating gas flow	: 7.0 L/min
DL temperature	: 200 °C
Desolvation temperature	: 450 °C
Interface voltage	: 3.0 kV
Qarray voltage	: 20/120V (for in-source CID)

3. Results

3-1. Analysis of pharmaceuticals

To investigate the effect of scan speed on chromatographic resolution, the pharmaceutical mixture sample was separated on UHPLC and MS acquired in cycles shown in Figure 1. Figures 2 and 3 compare the TIC (total ion current) chromatograms generated by different scan speed settings. With 15,000 u/sec scans, all 12 components could be resolved on the TIC chromatogram. With lower-speed scans, the chromatographic resolution was significantly compromised. Peak 2 became undetected, and the separation of Peaks 3 and 4 (positive mode) and Peaks 7 and 8 (negative mode) became insufficient.

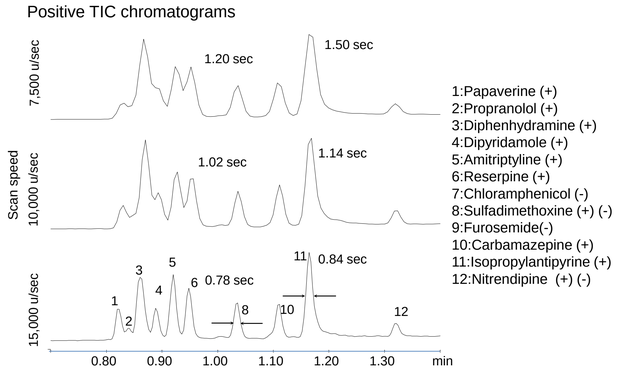

Figure 2. the positive TIC chromatograms acquired at different scan speeds

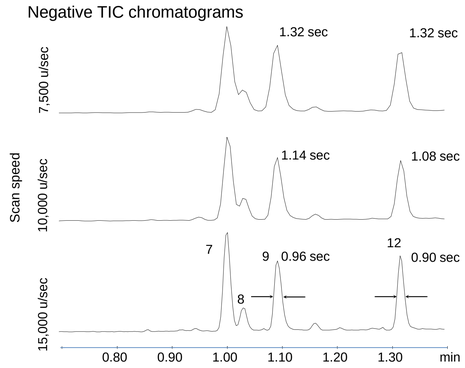

Figure 3. the negative TIC chromatograms in different scan speed

3-2. the peak widths in mass chromatogram

Figure 4 shows the extracted mass chromatograms of 12 pharmaceuticals acquired at 15,000 u/sec, and Table 3 summarizes the half widths of all peaks at different scan speed settings. The data made clear that a higher scan speed results in sharper peaks with smaller widths at half maximum.

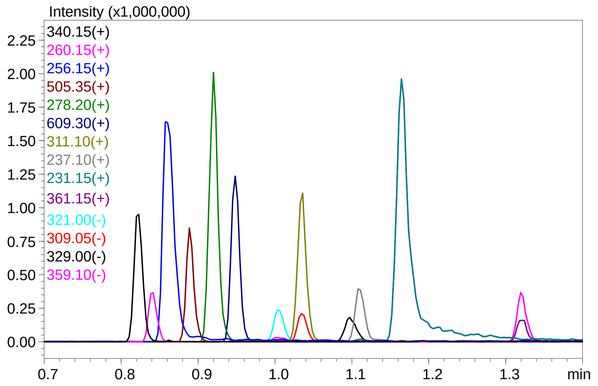

Figure 4. The mass chromatogram using 15,000 u/sec

Table 3. the peak widths at half maximum with different scan speeds

	Retention time (min)	the peak width at half height in 15,000 u/sec (sec)	the peak width at half height in 10,000 u/sec (sec)	the peak width at half height in 7,500 u/sec (sec)
1:papaverine (+)	0.834	0.78	1.02	1.20
2:Propranolol (+)	0.849	0.72	0.96	1.14
3:Diphenhydramine (+)	0.870	0.90	1.08	1.26
4:Dipyridamole (+)	0.896	0.78	1.02	1.20
5:Amitriptyline (+)	0.925	0.84	1.08	1.20
6:Recerpine (+)	0.953	0.72	1.02	1.14
8:Sulfadimethoxine (+)	1.042	0.78	1.08	1.26
10:Carbamazepine (+)	1.113	0.84	1.08	1.26
11:Isopropylantipyrine (+)	1.162	1.08	1.32	1.50
12:Nitrendipine (+)	1.325	0.84	1.02	1.20
7:Chloramphenicol (-)	1.004	0.72	0.96	1.20
8:Sulfadimethoxine (-)	1.038	0.72	1.02	1.20
9:Furosemide(-)	1.102	1.02	1.26	1.38
12:Nitrendipine (-)	1.317	0.84	1.08	1.26

3-3. the in-source CID results

Figure 5 shows analysis results of a normal scan spectrum and in-source CID spectrum and the structure and fragment site of reserpine. Using in-source CID as pseudo MS/MS, we can obtain the structural information of the compound.

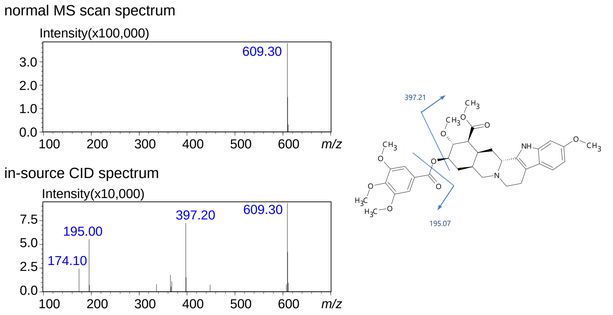

Figure 5. The mass spectra and structure and fragment sites of reserpine

4. Conclusion

- Ultrafast mass spectrometers are required to make full use of the high separation performance obtained by UHPLC. If the scan speeds are slow, peaks are detected broader, and the resolutions are lower.
- The modified LCMS-2020 enables to obtain a variety of MS data in a single analysis by measuring multiple modes within short acquisition times.