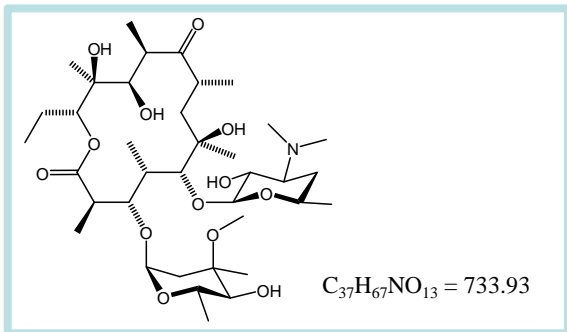


High-resolution and high-sensitivity are required for the analyses of related substances in drugs or impurities in chemicals. It is necessary to first achieve sufficient resolution in order to accurately measure the trace amount of a related substance or an impurity detected near the main component.

The high-resolution analysis in which erythromycin was analyzed as a model sample by Hitachi ultra high-speed liquid chromatograph, ChromasterUltra Rs, is introduced here as an analysis example of related substances in a drug. LaChrom II C18 for HPLC (4.6 mm I.D. × 150 mm, 5 μm) and LaChromUltra II C18 high resolution column for UHPLC (3.0 mm I.D. × 250 mm, 1.9 μm) were used and the results were compared.



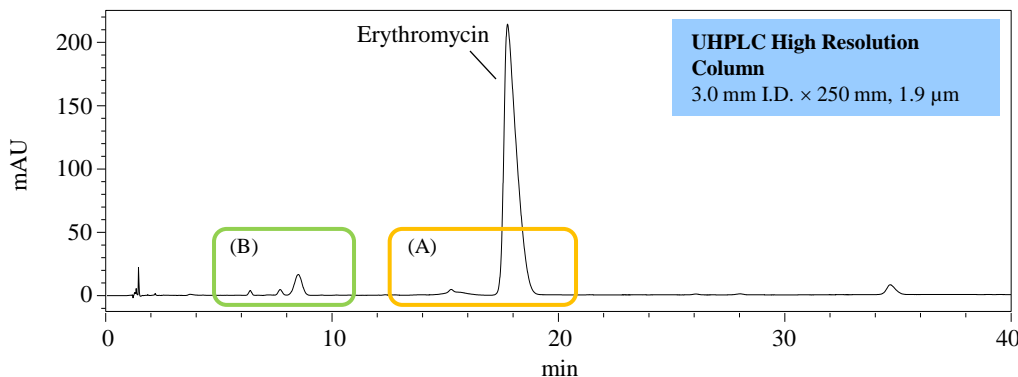
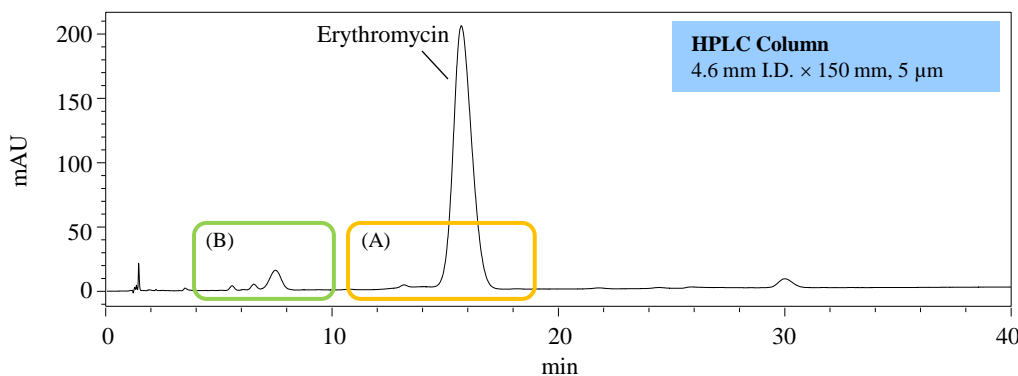
[Structural Formula of Erythromycin]



(Optional, Includes the parts prepared by a customer)

ChromasterUltra_{RS}

Analysis Example of Erythromycin



[Analysis Example of Erythromycin Standard Sample (10 mg/mL)]

When the erythromycin peaks were compared, the peak obtained by the UHPLC column was found to be sharper than the peak by the HPLC column. Very small peaks found near erythromycin (A) and the peaks found at about 4 - 12 min (B) are compared in the next page.

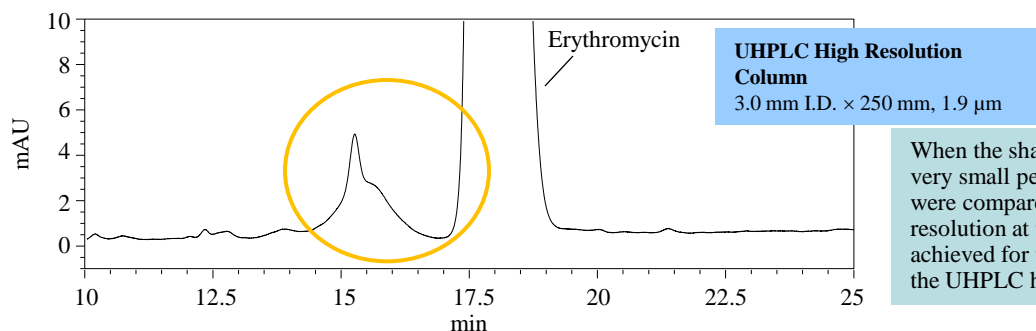
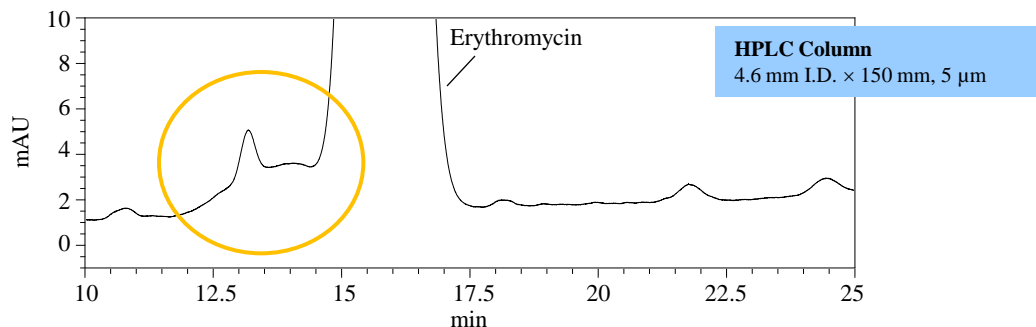
<Analytical Conditions for HPLC Column>

Column : LaChrom II C18 (5 μm) 4.6 mm I.D. × 150 mm
 Eluent : 20 mmol/L Phosphate buffer (pH 7.9) / Acetonitrile / Methanol = 45 / 40 / 15 (premix)
 Flow rate : 1.0 mL/min
 Column temperature : 50°C
 Detection wavelength : UV 210 nm (DAD)
 Injection vol. : 20 μL

<Analytical Conditions for UHPLC Column>

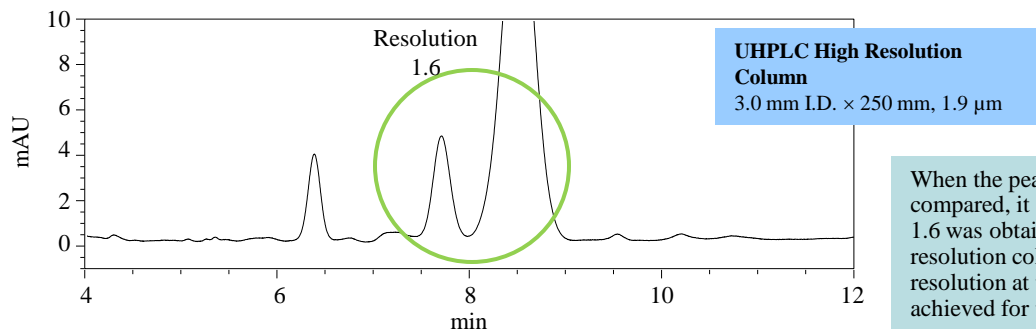
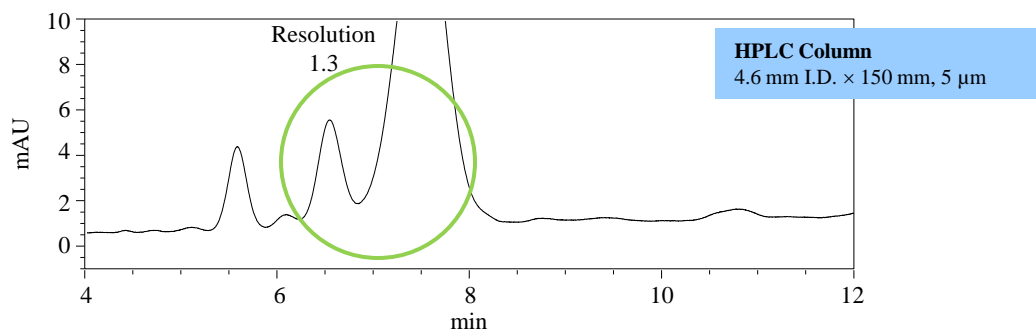
Column : LaChromUltra II C18 (1.9 μm) 3.0 mm I.D. × 250 mm
 Eluent : 20 mmol/L Phosphate buffer (pH 7.9) / Acetonitrile / Methanol = 45 / 40 / 15 (premix)
 Flow rate : 0.71 mL/min
 Column temperature : 50°C
 Detection wavelength : UV 210 nm (DAD)
 Injection vol. : 10 μL

■ Analysis Example of Erythromycin



When the shapes of erythromycin peak and a very small peak eluting before erythromycin were compared, it was found that the resolution at the baseline level can be achieved for the very small peak by using the UHPLC high resolution column.

[Analysis Example of Erythromycin Standard Sample (10 mg/mL), Enlarged for 10 - 25 min]



When the peaks eluting at 6 - 9 min were compared, it was found that the resolution of 1.6 was obtained by the UHPLC high resolution column, indicating that the resolution at the baseline level can be achieved for this peak as well.

[Analysis Example of Erythromycin Standard Sample (10 mg/mL), Enlarged for 4 - 12 min]

LaChromUltra II C18 column, with the adoption of inorganic-organic composite silica material having improved physical and chemical durability compared to the conventional silica gel, achieves high pressure resistance. As a result, a 250 mm high resolution column (1.9 μm) for UHPLC which can provide the number of theoretical plates of 50000/column was included in the lineup. For the analysis of related substances or impurities where many peaks emerge, good resolution can be achieved by using high resolution columns such as this one.

Main system configuration: ChromasterUltra Rs DAD system
(6170 Binary Pump, 6270 Autosampler, 6310 Column Oven, 6430 Diode Array Detector, Organizer)

NOTE: These data are an example of measurement; the individual values cannot be guaranteed.