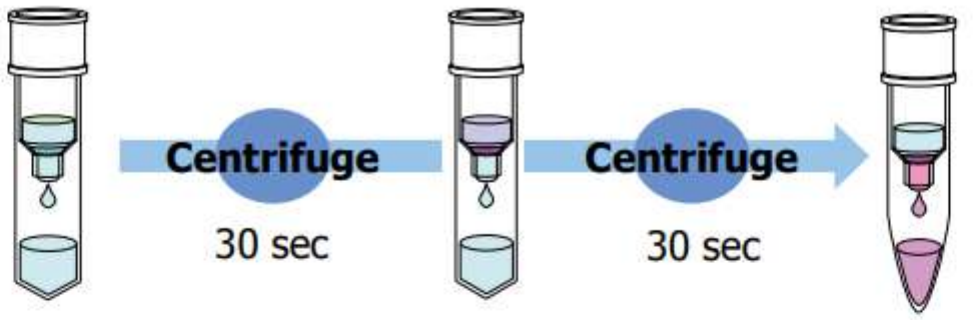
Sample Preparation

500 µL sample solution : First, desalt the peptide solution using MonoSpin C18. Then, dissolve the desalted sample solution with 0.1 % formic acd.

Centrifugation
Speed : 10,000 x g

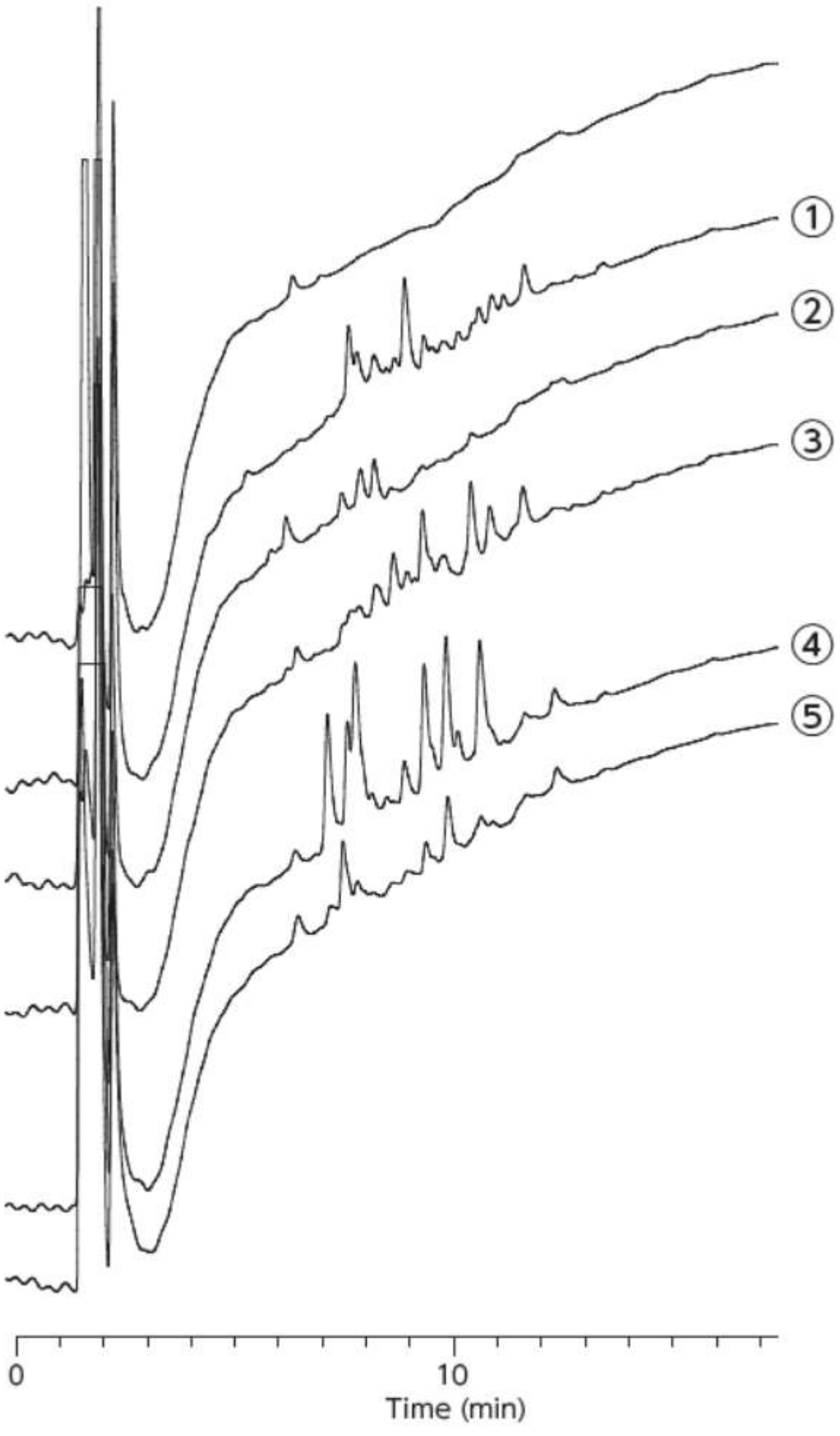


1. Conditioning
Add 300 µL 0.1 % formic acid

2. Adsorption
Add 500 µL peptide sample solution

3. Elution

Sample solution in 0.1 % formic acid



Always replace and attach a new recovery tube whenever adding a new elution buffer

Details of each elution buffer

① 25 mM $HCOONH_4$	200 µL
② 50 mM $HCOONH_4$	200 µL
③ 100 mM $HCOONH_4$	200 µL
④ 500 mM $HCOONH_4$	200 µL
⑤ 1 M $HCOONH_4$	200 µL

* Each elution buffer contains 10 % acetonitrile

Conditions

Column : Inertsil ODS-3 (3 µm, 2.1 x 150 mm)
Eluent : A) H_2O (0.1 % HCOOH)
B) Acetonitrile (0.1 % HCOOH)
A/B = 90/10 - 20 min - 50/50
Detection : UV 210 nm
Flow Rate : 0.2 mL/min
Col. Temp. : 40 °C
Injection Vol. : 2 µL