Direct Injection of Blood Plasma for the Determination of Drugs using "Co-Sense for BA" (Part 4)

The "Co-Sense for BA" is a biosample analysis system capable of directly injecting biosamples such as blood plasma and blood serum. The "Co-Sense for BA" system removes proteins contained in the sample from the pretreatment flow path. The target components are extracted in the pretreatment column, eluted into the analysis flow path, separated from matrix components, and detected.

The principle and application examples of the Co-Sense for BA were already introduced in Application News L285, L286, and L293. This Application News introduces analysis examples using mobile phases with neutral pH. The analyses were carried out injecting control human blood plasma that was filtered through a 0.45µm membrane filter and spiked with

■ Determination of Phenylbutazone in Blood Plasma

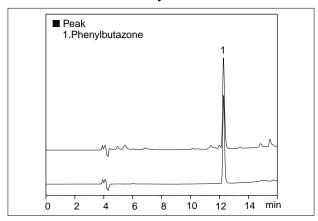


Fig.1 Chromatogram of Phenylbutazone in Blood Plasma (upper:spiked 4μg/mL, 50μL injected; lower:4μg/mL standard, 50μL injected)

Table 1 Analytical Conditions

For Sample Injection

: Shim-pack MAYI-ODS(10mmL.×4.6mmI.D.) Column

Mobile Phase: A: 100mM Acetate (Na) buffer (pH4.7)

B: Acetonitrile

A / B = 95 / 5 (v/v)

Flow Rate : 2.0mL/min Dilution Factor · 8

For Separation

Column : Shim-pack FC-ODS (75mmL.×4.6mmI.D.)

Mobile Phase : A: 20mM Phosphate (Na) buffer (pH=6.9)

100mM Sodium perchlorate B: Methanol

Linear gradient B 30%→80%

 $(4\min \rightarrow 12\min)$

Flow Rate : 1.0mL/min

Temperature : 40°C

Detection : SPD-M10AvP at 265nm

Determination of Ibuprofen in Blood Plasma

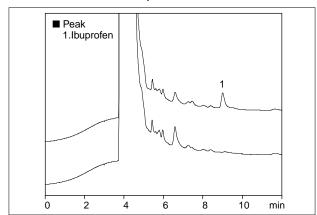


Fig.2 Chromatogram of Ibuprofen in Blood Plasma (upper:spiked 1µg/mL, 50µL injected; lower:unspiked, 50µL injected)

Table 2 Analytical Conditions

For Sample Injection

: Shim-pack MAYI-ODS(10mmL.×4.6mmI.D.) Column

Mobile Phase : A: 100mM Acetate (Na) buffer (pH4.7)

B: Acetonitrile

A / B = 90 / 10 (v/v): 2.0mL/min

Flow Rate Dilution Factor: 8

For Separation

Column : Shim-pack FC-ODS (75mmL.×4.6mmI.D.) Mobile Phase: A: 20mM Phosphate (Na) buffer (pH=6.9)

B. Methanol

A / B = 45 / 55 (v/v)

Flow Rate : 1.0mL/min : 40°C

Temperature Detection : SPD-M10AvP at 210nm

■ Determination of Indometacin in Blood Plasma

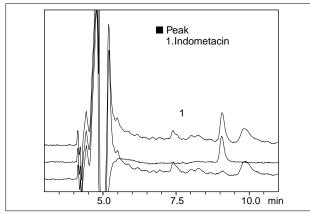


Fig.3 Chromatogram of Indometacin in Blood Plasma (upper:spiked 100ng/mL, 50μL injected; middle:100ng/mL standard, 50μL injected; lower:unspiked, 50μL injected)

Table 3 Analytical Conditions

For Sample Injection

Column : Shim-pack MAYI-ODS(10mmL.×4.6mmI.D.)

Mobile Phase: A: 100mM Ammonium acetate

B: Acetonitrile

 $A \, / \, B = 90 \, / \, 10 \; (v/v) \label{eq:abs}$ Flow Rate : 4.0 mL/min

Dilution Factor: 8 For Separation

> Column : Shim-pack VP-ODS (150mmL.×4.6mmI.D.) Mobile Phase : A: 20mM Phosphate (Na) buffer (pH=6.9)

B: Acetonitrile

Linear gradient B 30%→35%

(4min→6min)

 $\begin{array}{ll} \hbox{Flow Rate} & : 1.2 mL/min \\ \hbox{Temperature} & : 40 ^{\circ} C \end{array}$

Detection : SPD-M10AvP at 270nm

■ Determination of Lidocaine in Blood Plasma

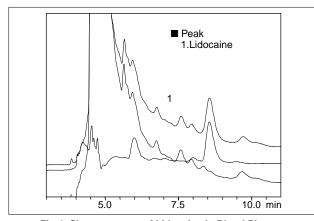


Fig.4 Chromatogram of Lidocaine in Blood Plasma (upper:spiked 1μg/mL, 50μL injected; middle:1μg/mL standard, 50μL injected; lower:unspiked, 50μL injected)

Table 4 Analytical Conditions

For Sample Injection

Column : Shim-pack MAYI-ODS(10mmL.×4.6mmI.D.)

Mobile Phase : A: 50mM Phosphate (Na) buffer (pH=6.9)

B: Acetonitrile

 $A \, / \, B = 90 \, / \, 10 \; (v/v) \label{eq:approx}$ Flow Rate : 4.0 mL/min

Dilution Factor : 8 For Separation

Column : Shim-pack VP-ODS (150mmL.×4.6mmI.D.)
Mobile Phase : A: 20mM Phosphate (Na) buffer (pH=6.9)

B: Acetonitrile

A / B = 45 / 55 (v/v): 1.2mL/min

Flow Rate : 1.2mL/ Temperature : 40°C

Detection : SPD-M10AvP at 220nm

■ Determination of Triazoram in Blood Plasma

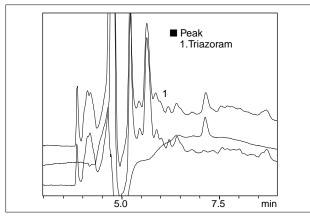


Fig.5 Chromatogram of Triazoram in Blood Plasma (upper:spiked 20ng/mL, 200μL injected; middle:20ng/mL standard, 200μL injected; lower:unspiked, 200μL injected)

Table 5 Analytical Conditions

For Sample Injection

Column : Shim-pack MAYI-ODS(10mmL.×4.6mmI.D.)

Mobile Phase : A: 20mM Ammonium acetate

B: Acetonitrile A / B = 90 / 10 (v/v)

Flow Rate : 4.0mL/min Dilution Factor : 8

For Separation

Column : Shim-pack VP-ODS (150mmL.×4.6mmI.D.)

Mobile Phase : A: 10mM Ammonium acetate

B: Acetonitrile

Linear gradient B 45%→50%

 $(4min\rightarrow 6min)$

Flow Rate : 1.2mL/min

Temperature : 40°C

Detection : SPD-M10A_{VP} at 250nm

*Data presented here was not acquired using instruments approved under the Japanese Pharmaceutical Affaires Law



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