

# HPLC Application

ID No.: 25490

## Extraction of Opiates in Human Serum Using Strata-X PRO and a Kinetex 2.6 µm Biphenyl LC Column

**Column:** Kinetex<sup>®</sup> 2.6 µm Biphenyl 100 Å, LC Column 50 x 3.0 mm, Ea

**Dimensions:** 50 x 3 mm ID

**Order No:** 00B-4622-Y0

**Elution Type:** Gradient

**Eluent A:** 0.1% Formic acid in Water

**Eluent B:** 0.1% Formic acid in Methanol

Gradient Profile:	Step No.	Time (min)	Pct A	Pct B
	1	0	90	5
	2	4	5	95
	3	5.5	5	95
	4	5.51	95	5
	5	7	95	5

**Flow Rate:** 0.5 mL/min

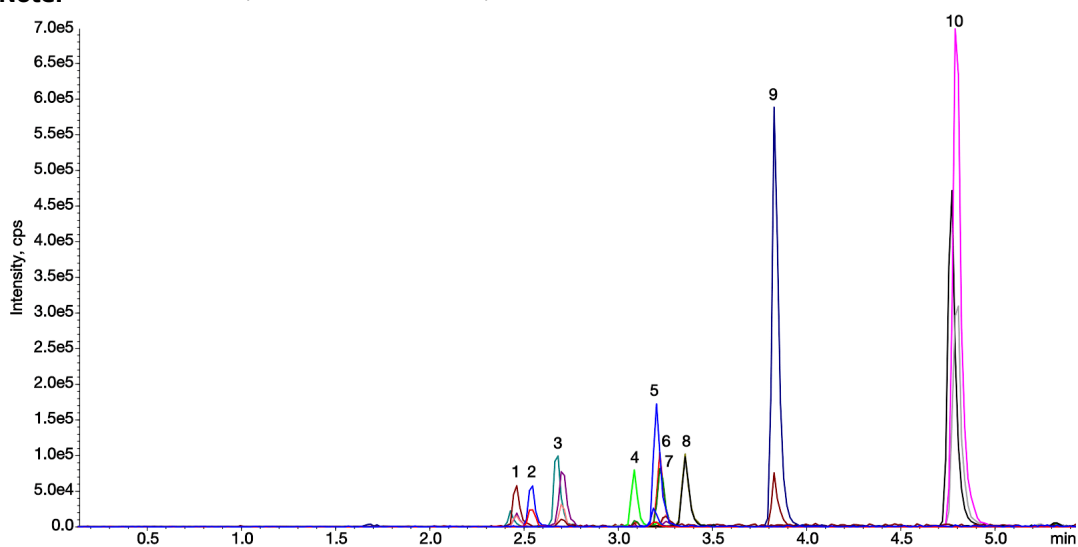
**Col. Temp.:** 45 °C

**Detection:** LC/MS/MS @ (ambient)

**Analyst Note:** Human Serum Analyte diluted with initial mobile phase



Products used in this application:



## Extraction of Opiates in Human Serum Using Strata-X PRO and a Kinetex 2.6 $\mu$ m Biphenyl LC Column

### ANALYTES:

- 1 Morphine
- 2 Oxymorphone
- 3 Hydromorphone
- 4 Naloxone
- 5 6-MAM
- 6 Codeine
- 7 Oxycodone
- 8 Hydrocodone
- 9 Norfentanyl
- 10 Fentanyl



# Sample Preparation Details

for HPLC Application ID No.: 25490

## Extraction of Opiates in Human Serum Using Strata-X PRO and a Kinetex 2.6 µm Biphenyl LC Column

### PRODUCT DESCRIPTION:

Order No.:

### SOLID PHASE EXTRACTION (SPE) PROCEDURE:

**Note:** The solvent volumes shown below are for a bed mass.

The solvent volumes will need to be adjusted for a smaller or larger bed mass.

#### Condition:

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#### Load:

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#### Wash:

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#### Dry:

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#### Elute:

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#### Final Prep and Analysis:

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200 µL Human serum diluted 1:1 with 1% Formic acid in Water

Inject: 10 µL on HPLC LC/MS/MS @ (ambient)

<b>ANALYTES:</b>	<b>Spiked Conc. (ng/mL)</b>	<b>Log P</b>	<b>pKa</b>	<b>% Rec</b>	<b>%RSC (n=0)</b>
<b>1</b> Morphine	50			68	
<b>2</b> Oxymorphone	50			80	
<b>3</b> Hydromorphone	50			75	
<b>4</b> Naloxone	50			83	
<b>5</b> 6-MAM	50			77	
<b>6</b> Codeine	50			70	
<b>7</b> Oxycodone	50			64	
<b>8</b> Hydrocodone	50			73	
<b>9</b> Norfentanyl	50			57	
<b>10</b> Fentanyl	50			79	

**Note:** This method is designed as a convenient starting point for further investigation and can be tailored to meet your extraction goals. Call your local Phenomenex Representative for assistance in method development and optimization techniques.

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