

## Validated Analytical Methods

<b>COMPOUND</b>	<b>Sample pretreatment</b>	<b>Separation</b>	<b>Detection</b>	<b>Matrix</b>
<b>CAFFEINE</b>	Protein precipitation	UPLC	MS	Plasma Microsomes
<b>CEFEPIME</b>	Ultrafiltration	HPLC	MS	Plasma Urine
<b>CELECOXIB</b>	Protein precipitation	HPLC	MS	Plasma
<b>CIPROFLOXACINE</b>	Ultrafiltration Liquid-Liquid Extraction	HPLC	Fluorescence	Plasma Urine
<b>CISPLATIN</b>	Protein precipitation	UPLC	MS	Plasma
<b>CURCUMA</b>	Liquid-Liquid Extraction	HPLC	MS	Plasma
<b>DEXMEDETOMIDINE</b>	Protein precipitation	UPLC	MS	Plasma
<b>EFAVIRENZ</b>	Liquid-Liquid Extraction	HPLC	MS	Microsomes
<b>ENTACAPONE</b>	Liquid-Liquid Extraction	HPLC	MS	Microsomes
<b>FEBANTEL</b>	Liquid-Liquid Extraction	HPLC	MS	Plasma
<b>FLUBENDAZOLE</b>	Liquid-Liquid Extraction	HPLC	MS	Plasma
<b>GALANTAMINE</b>	Liquid-Liquid Extraction	HPLC	MS	Plasma Brain tissue
<b>GLYCOPYRROLATE</b>	Liquid-Liquid Extraction	HPLC	MS	Plasma

<b>ITRACONAZOLE</b>	Liquid–Liquid Extraction	HPLC	MS	Plasma
<b>METOPROLOL</b>	Liquid–Liquid Extraction	HPLC	MS	Plasma
<b>OFLOXACINE - MOXIFLOXACINE</b>	Protein precipitation	HPLC	MS	Plasma
<b>PACLITAXEL</b>	SPE	UPLC	MS	Rat tumor tissue
<b>PROPOFOL AND METABOLITES</b>	SPE	UPLC	MS	Plasma Microsomes
<b>PYRIMETHAMINE</b>	Protein precipitation	HPLC	MS	Plasma
<b>REMIFENTANIL</b>	Protein precipitation	UPLC	MS	Plasma
<b>SULFALENE</b>	Protein precipitation	HPLC	MS	Plasma
<b>THEOPHYLLINE</b>	Protein precipitation	UPLC	MS	Plasma Microsomes

- **Caffeine and its metabolites (Theophylline, Paraxanthin, Theobromine)**  
The samples which can be analyzed are plasma and microsomes. The extraction method we use is a protein precipitation. The samples are analyzed on an ultra-high-performance liquid chromatography-triple quadrupole mass spectrometry (Acquity UPLC – Ultima with “Multiple Reaction Monitoring” (MRM)).
- **Cefepime**  
Cefepime is measured in plasma and urine samples. The samples are extracted with ultrafiltration. They are analyzed on a high-performance liquid chromatography-triple quadrupole mass spectrometry (Agilent 1100 – API3000 with “Multiple Reaction Monitoring” (MRM)).
- **Celecoxib**

Celecoxib concentrations are measured in plasma using a protein precipitation. The samples are analyzed on a high-performance liquid chromatography-triple quadrupole mass spectrometry (Agilent 1100 – API3000 with “Multiple Reaction Monitoring” (MRM)).

- **Ciprofloxacin**

Ciprofloxacin is quantitatively measured in plasma and urine samples. In addition ultrafiltration can be used to measure the unbound (to protein) concentration. The analysis is performed using high-performance liquid chromatography with fluorescence detection. First a protein precipitation is performed.

- **Cisplatin**

Cisplatin concentrations in plasma samples are measured using ultra-high-performance liquid chromatography-triple quadrupole mass spectrometry (Acquity UPLC H-class Xevo TQS with “Multiple Reaction Monitoring” (MRM)). The extraction method is a protein precipitation.

- **Curcuma**

Curcuma is measured in plasma samples. The samples are analyzed on a high-performance liquid chromatography-triple quadrupole mass spectrometry (Agilent 1100 – API3000 with “Multiple Reaction Monitoring” (MRM)). The sample treatment is a liquid-liquid extraction.

- **Dexmedetomidine:**

Dexmedetomidine concentrations in plasma samples are measured using ultra-high-performance liquid chromatography-triple quadrupole mass spectrometry (Acquity UPLC H-class Xevo TQS with “Multiple Reaction Monitoring” (MRM)). The extraction method is a protein precipitation.

- **Efaverinz**

Efaverinz can be measured in human liver microsomes. The sample extraction method is a liquid-liquid extraction. Analysis is on a high-performance liquid chromatography-triple quadrupole mass spectrometry (Agilent 1100 – API3000 with “Multiple Reaction Monitoring” (MRM)).

- **Entacapone –tolcapone**  
Tol- and entacapone can be measured in human liver microsomes. The sample extraction method is a liquid-liquid extraction. Analysis is on a high-performance liquid chromatography-triple quadrupole mass spectrometry (Agilent 1100 – API3000 with “Multiple Reaction Monitoring” (MRM)).
- **Febantel**  
Febantel concentrations are measured in plasma samples. A high-performance liquid chromatography-triple quadrupole mass spectrometry (Agilent 1100 – API3000 with “Multiple Reaction Monitoring” (MRM)) system is used for the analytical phase. The sample pretreatment is liquid-liquid extraction.
- **Flubendazole**  
Flubendazole and its metabolites (2-amino flubendazole and hydroxy flubendazole) can quantitatively be measured in plasma samples. Analysis is on a high-performance liquid chromatography-triple quadrupole mass spectrometry (Agilent 1100 – API3000 with “Multiple Reaction Monitoring” (MRM)) system. The sample treatment is a liquid-liquid extraction.
- **Galantamine**  
Galantamine can be measured in plasma and brain tissue. The sample extraction method is liquid-liquid extraction. Analysis is performed on a high-performance liquid chromatography-triple quadrupole mass spectrometry (Agilent 1100 – API3000 with “Multiple Reaction Monitoring” (MRM)).
- **Glycopyrrolate**  
Glycopyrrolate is measured in human plasma. The sample pretreatment is liquid-liquid extraction. Analysis is performed on a ultra-high-performance liquid chromatography-triple quadrupole mass spectrometer.
- **Itraconazole**  
Itraconazole is measured in plasma samples. It is analyzed using a high-performance liquid chromatography-triple quadrupole mass spectrometry (Agilent 1100 – API3000 with “Multiple Reaction Monitoring” (MRM)). The sample treatment is liquid-liquid extraction.
- **Metoprolol**

Metoprolol is measured in human plasma .

The sample pretreatment is liquid-liquid extraction.

Analysis is performed using a high-performance liquid chromatography-triple quadrupole mass spectrometer.

- **Ofloxacin - Moxifloxacin**

Ofloxacin and Moxifloxacin concentrations can be measured in plasma samples. The analysis is performed using a high-performance liquid chromatograph with fluorescence detection.(Waters 2695 HPLC – Waters470 Fluorescence detector). First a protein precipitation is performed.

- **Paclitaxel**

Paclitaxel is measured in rat tissue (rat tumor). The samples are extracted with SPE and analyzed using a ultra-high-performance liquid chromatography-triple quadrupole mass spectrometry (Acquity UPLC – Ultima with “Multiple Reaction Monitoring” (MRM)).

- **Propofol and metabolites**

Propofol and metabolites (propofol glucuronide (PG), 4-hydroxy-propofol (4HP), 2,6 Diisopropyl-1,4-benzoquinone (Imp J), 1-quinol glucuronide (1QG), and 4-quinol sulphate (4QS) are simultaneously measured in human plasma samples or in microsomal matrix. The extraction procedure is solid phase (SPE). The analysis is performed on a ultra-high-performance liquid chromatography-triple quadrupole mass spectrometer (Acquity UPLC-Ultima with Multiple Reaction Monitoring (MRM)).

- **Pyrimethamine and Sulfalene**

PYR/SUL are simultaneously measured in human plasma. Sample clean-up is achieved by protein precipitation. The chromatographic system consisted of an Acquity Separation Module. Detection was performed using a Quattro Ultima triple quadrupole instrument.

- **Remifentanyl:**

Remifentanyl concentrations in plasma samples are measured using ultra-high-performance liquid chromatography- triple quadrupole mass spectrometry (Acquity UPLC H-class Xevo TQS with “Multiple Reaction Monitoring” (MRM)). The extraction method is protein precipitation.