

HPLC Application 0008

Benzodiazepine LC-MS/MS Screening

Column comparison: Dr. Maisch HPLC GmbH
ReproSil Star Phenyl-Ether 3 μm , 50 mm x 2.0 mm
(PN: star3.pet.s0502)

vs.

Phenomenex
Synergi Polar RP 4 μm , 50 mm x 2.1 mm
(PN: 00B-4336-AN)

In this application 2 similar modified phenyl-phases (fully-porous silica modified with phenyl-ether & polar end-capping) were compared for the screening of different benzodiazepines with standards and different real blood samples.

ID No.: 0008

Gradient Method

Time program for gradient (binary high-pressure pumps)

	Mobile Phase B (%)	Flow	Comments
Minute		$\mu\text{L}/\text{min}$	
0.00	10	300	Start MS and pumps
1.00	10	300	Start gradient
7.50	90	300	
8.50	90	300	
8.60	10	300	
12.00	10	300	End of program

Eluent A:	H ₂ O with 0.1 % formic acid
Eluent B:	MeCN with 0.1 % formic acid
Flow Rate:	300 mL/min
Temperature:	30°C
Injection:	10 µL
MS Parameters:	Sciex 3200 QTrap in MRM mode (MS/MS) Positive electrospray ionization, Spray voltage: 4500 V and temperature at 450°C Gas 1: 40, Gas 2: 60, Curtain gas: 40, Collision Gas: 6 MS/MS conditions tuned for each compound separately

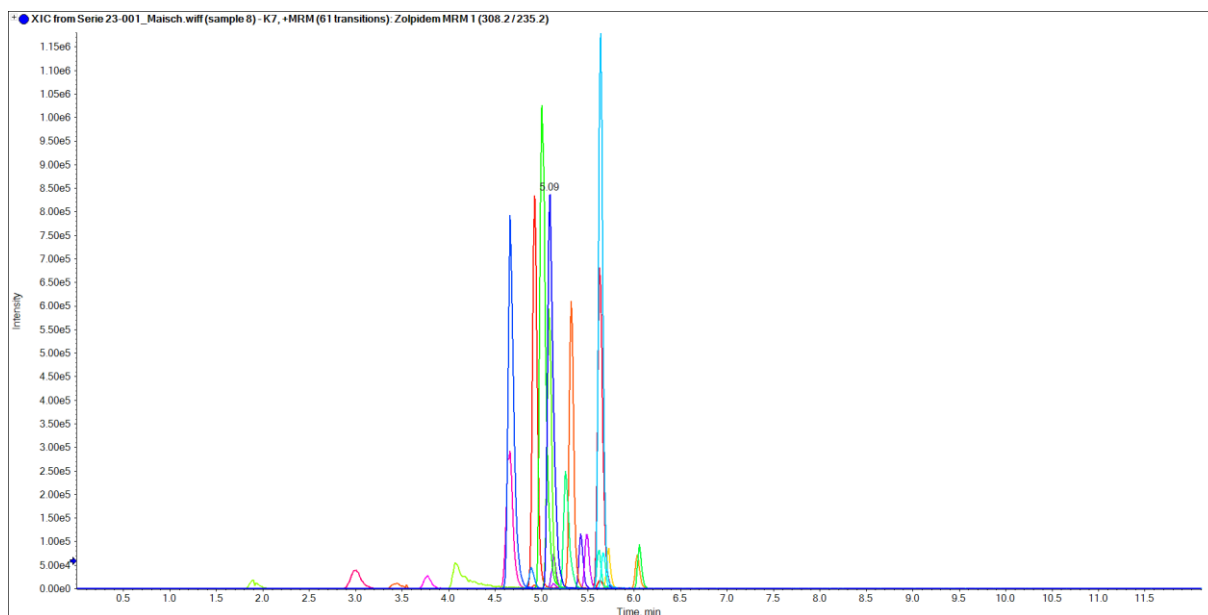
Separation of benzodiazepins:

- Clonazepam Klonopin^{*)}, Ravotril^{*)}
- Norflunitrazepam Desmethylflunitrazepam^{**)}, N-Desmethylflunitrazepam^{**) flunitrazepam metabolite}
- Temazepam Restoril^{*)}
- Zopiclone Imovane^{*)}
- Nordiazepam Nordazepam, Desoxydemoxepam, Desmethyldiazepam^{**) Nordaz^{*)}, Stilny^{*)}, Madar^{*)}, Vegesan^{*)}, and Calmday^{*)}}
- 7-NH₂-Flunitrazepam flunitrazepam metabolite
- Diazepam Valium^{*)}
- Oxazepam Alepam^{*)}
- Desalkylfluorazepam Norflurazepam^{**)}
- Aprazolam Xanax^{*)}
- Flunitrazepam Rohypnol^{**)}
- Bromazepam Lexotanil^{*)}
- Lorazepam Ativan^{*)}
- Midazolam Versed^{*)}
- Triazolam Halcion^{*)}
- Flurazepam Dalmane^{*)} Dalmadorm^{*)}
- Lormetazepam Noctamid^{*)}
- Zaleplon Sonata^{*)}
- Clobazam Frisium^{*)} Onfi^{*)}
- Nitrazepam Mogadon
- Alpha-hydroxymidazolam Midazolam metabolite

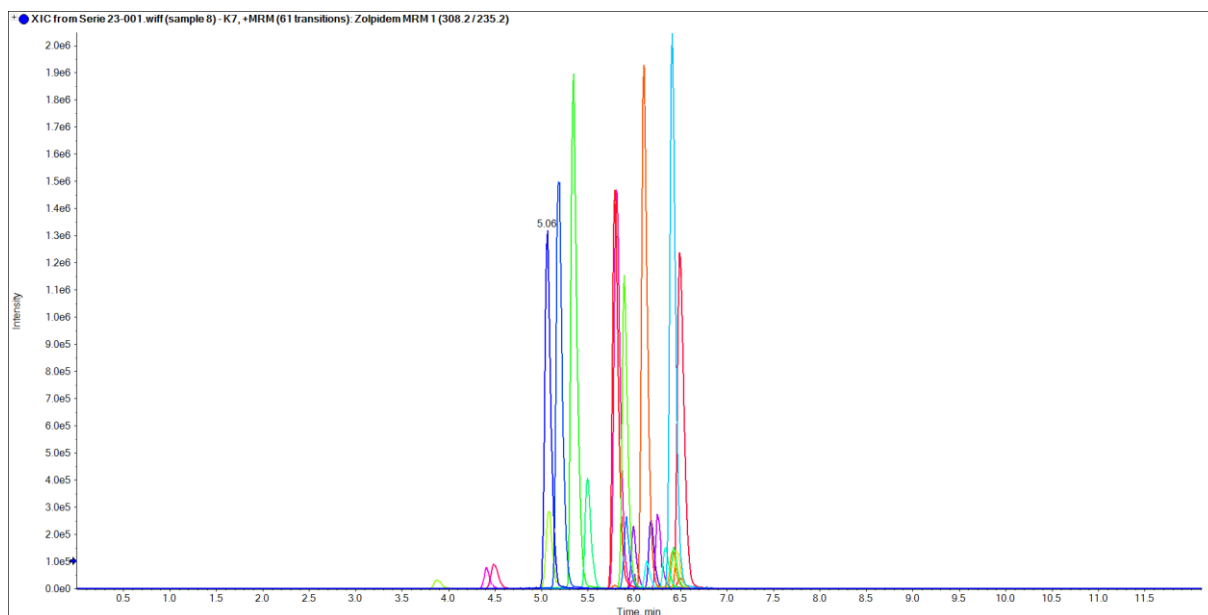
^{*)} Brand name ^{**) also known as}

K7-Standards (Benzodiazepine Mix)

ReproSil Star Phenyl-Ether - Dr. Maisch HPLC GmbH

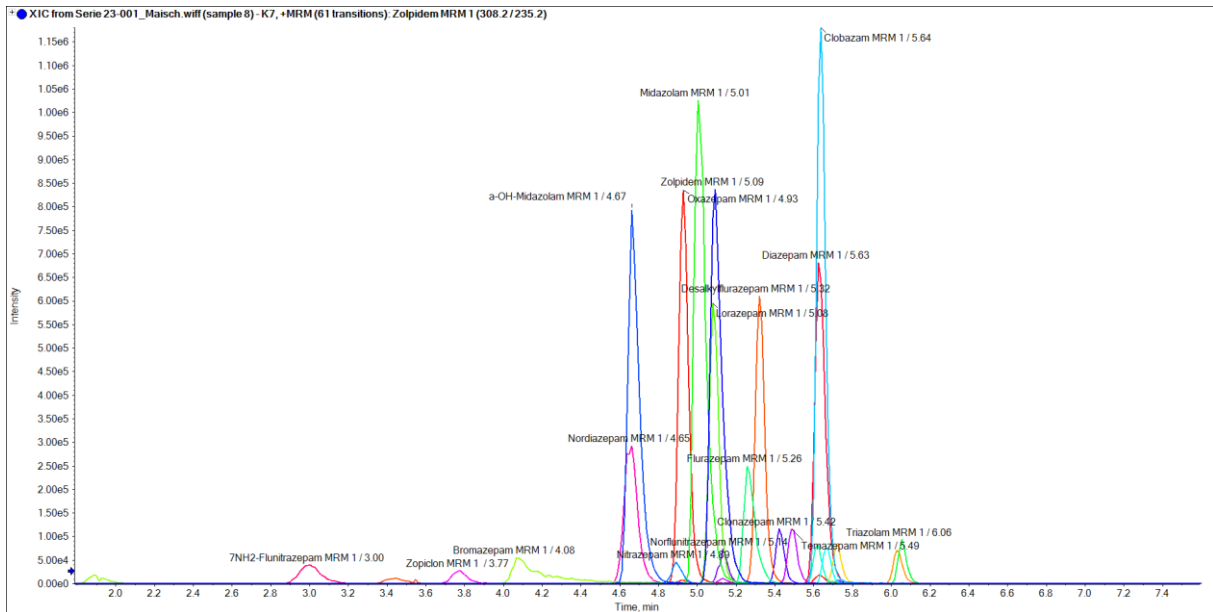


Synergi Polar RP - Phenomenex

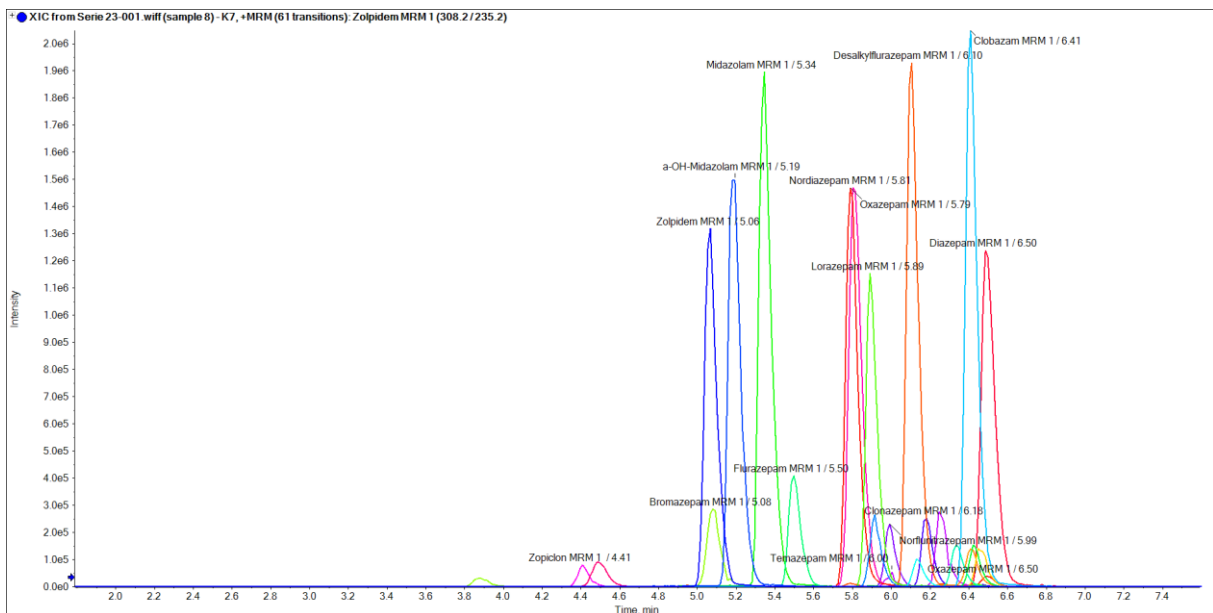


K7-Standards (Benzodiazepine Mix) – ZOOM 1

ReproSil Star Phenyl-Ether - Dr. Maisch HPLC GmbH

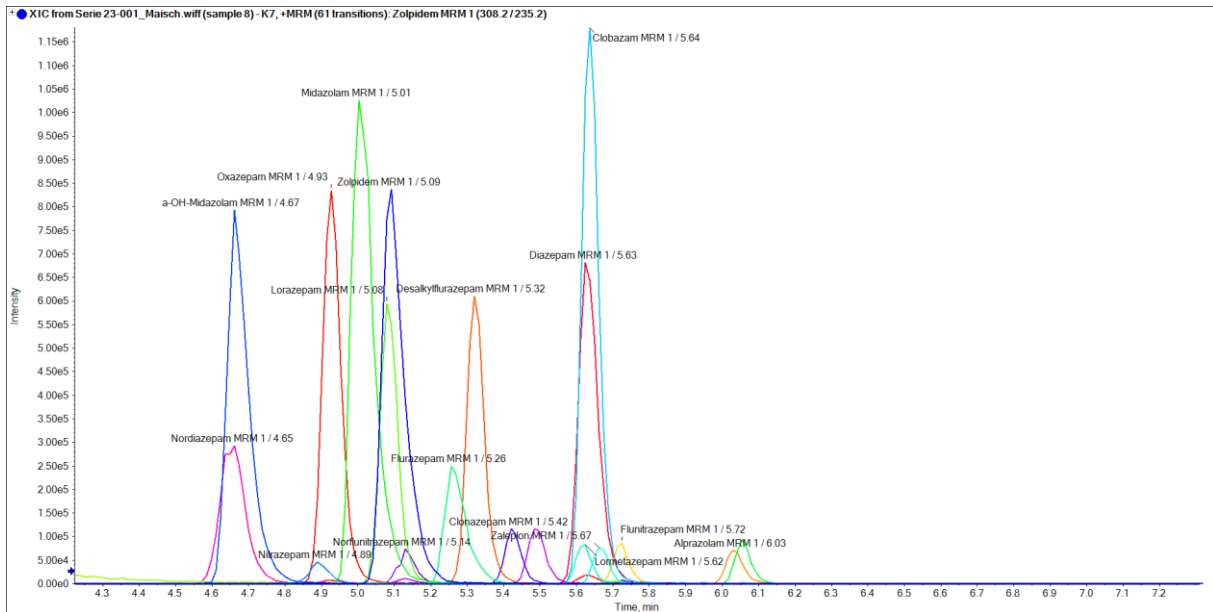


Synergi Polar RP - Phenomenex

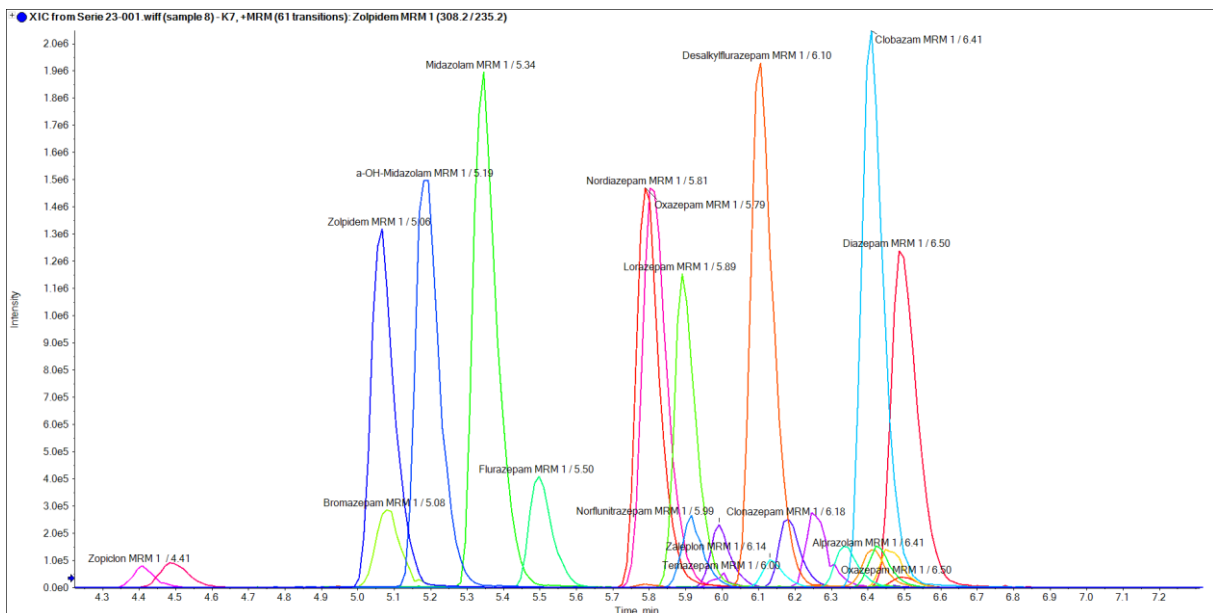


K7-Standards (Benzodiazepine Mix) – ZOOM 2

ReproSil Star Phenyl-Ether - Dr. Maisch HPLC GmbH

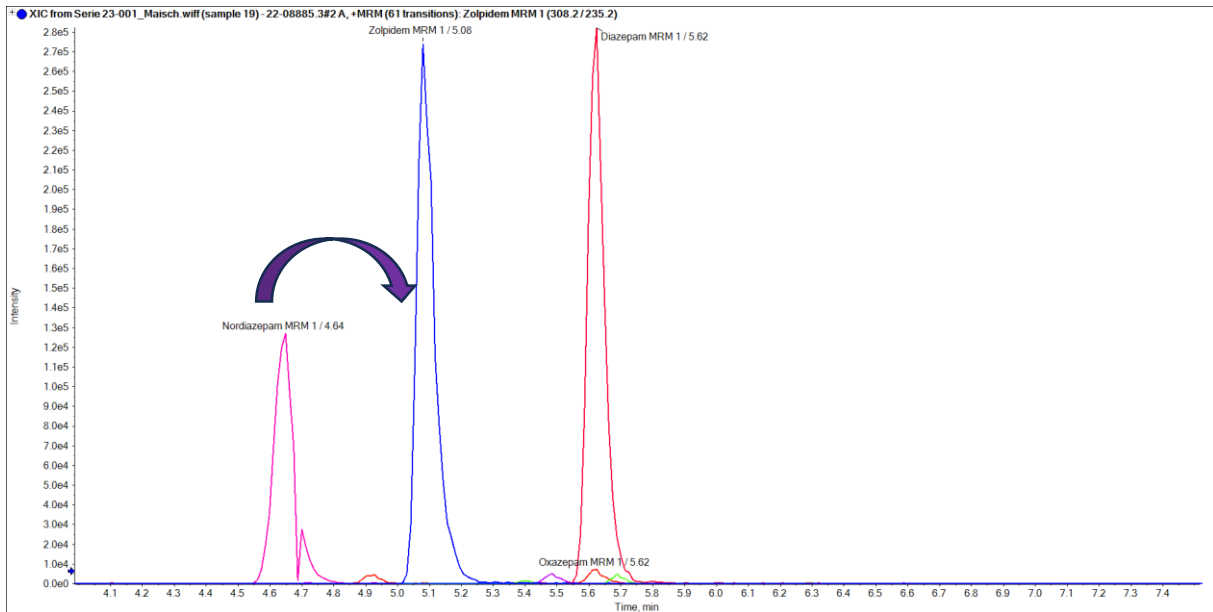


Synergi Polar RP - Phenomenex

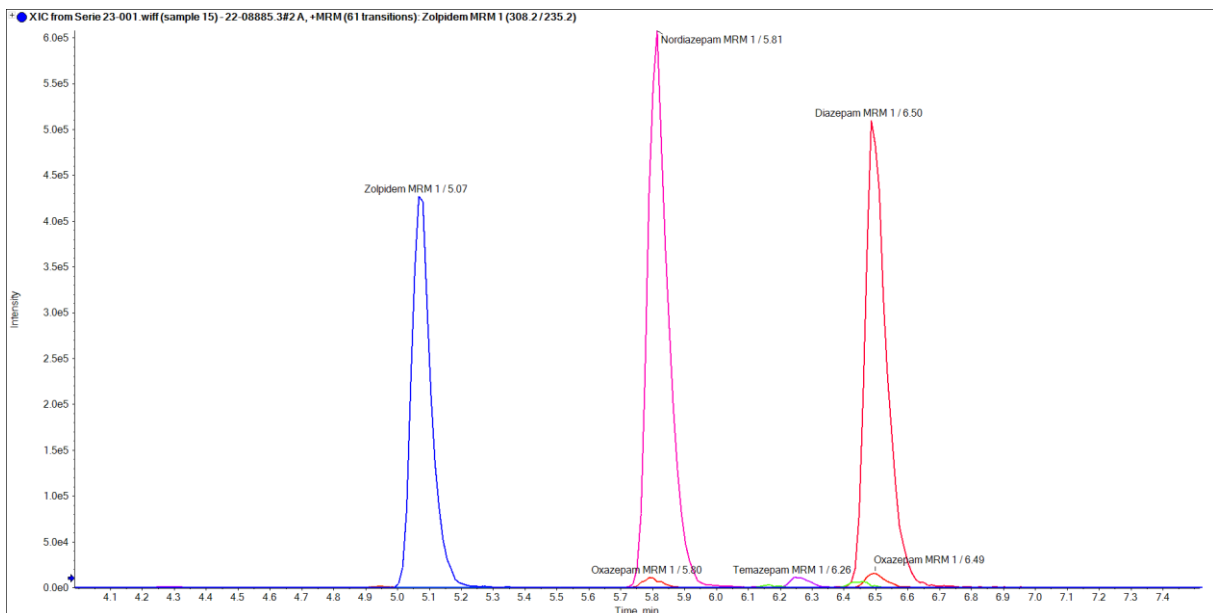


Blood sample 19

ReproSil Star Phenyl-Ether - Dr. Maisch HPLC GmbH

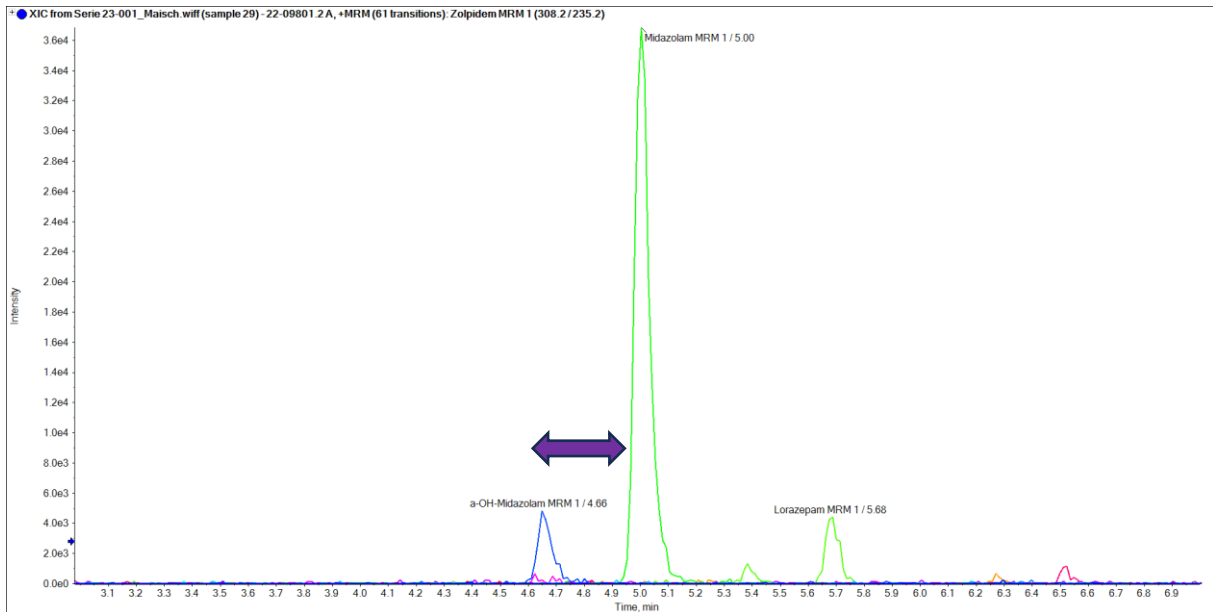


Synergi Polar RP - Phenomenex

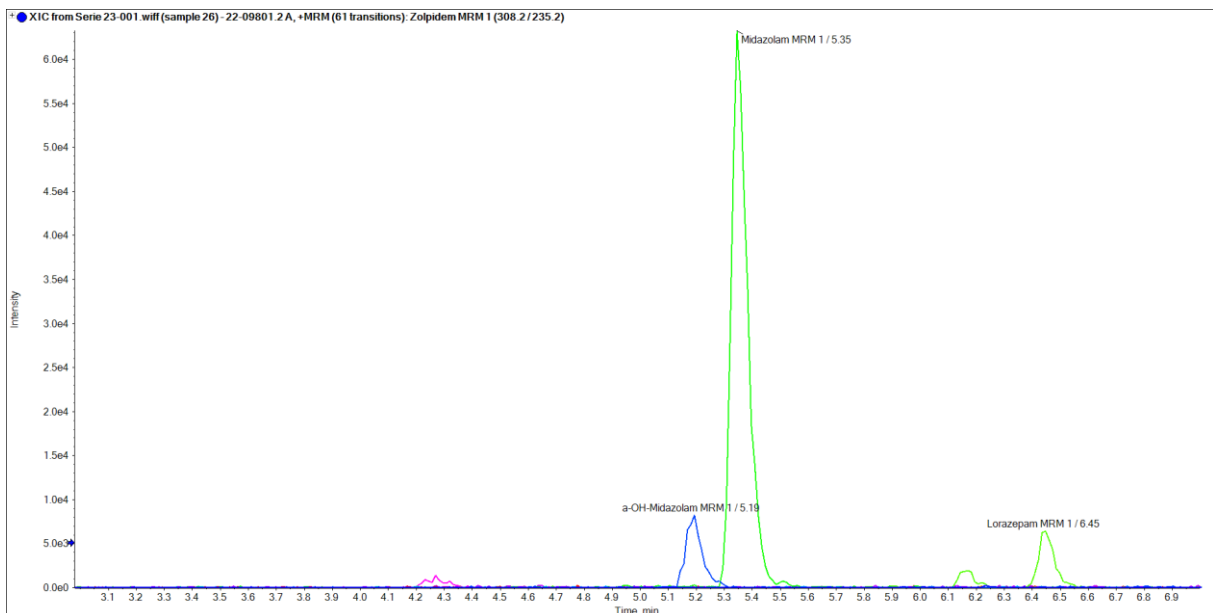


Blood sample 29

ReproSil Star Phenyl-Ether - Dr. Maisch HPLC GmbH

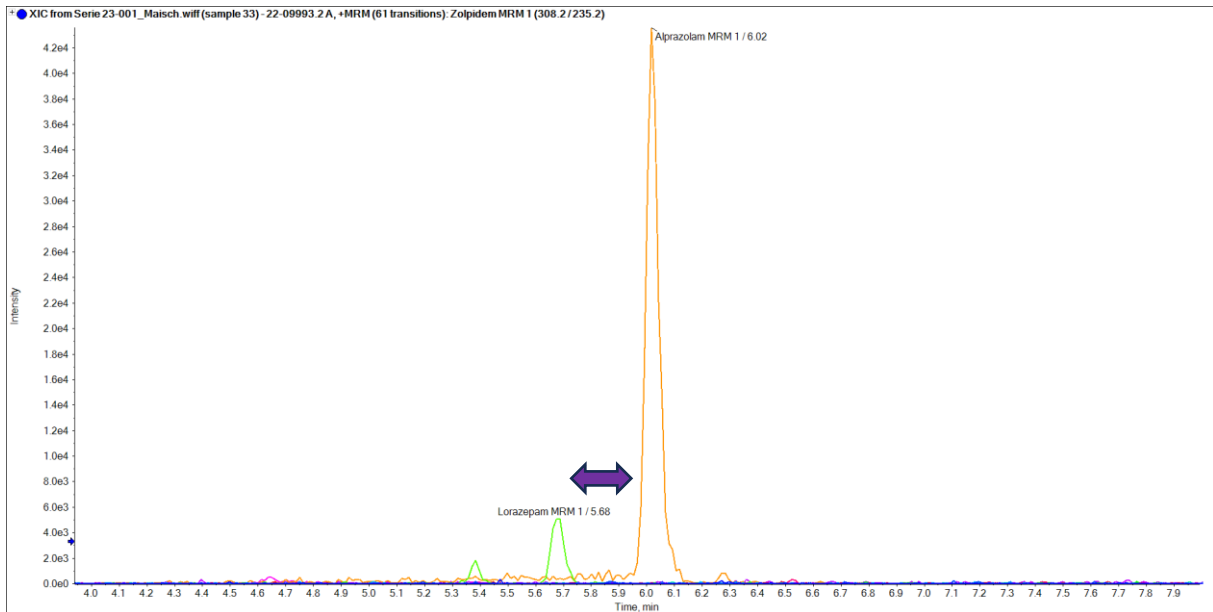


Synergi Polar RP - Phenomenex

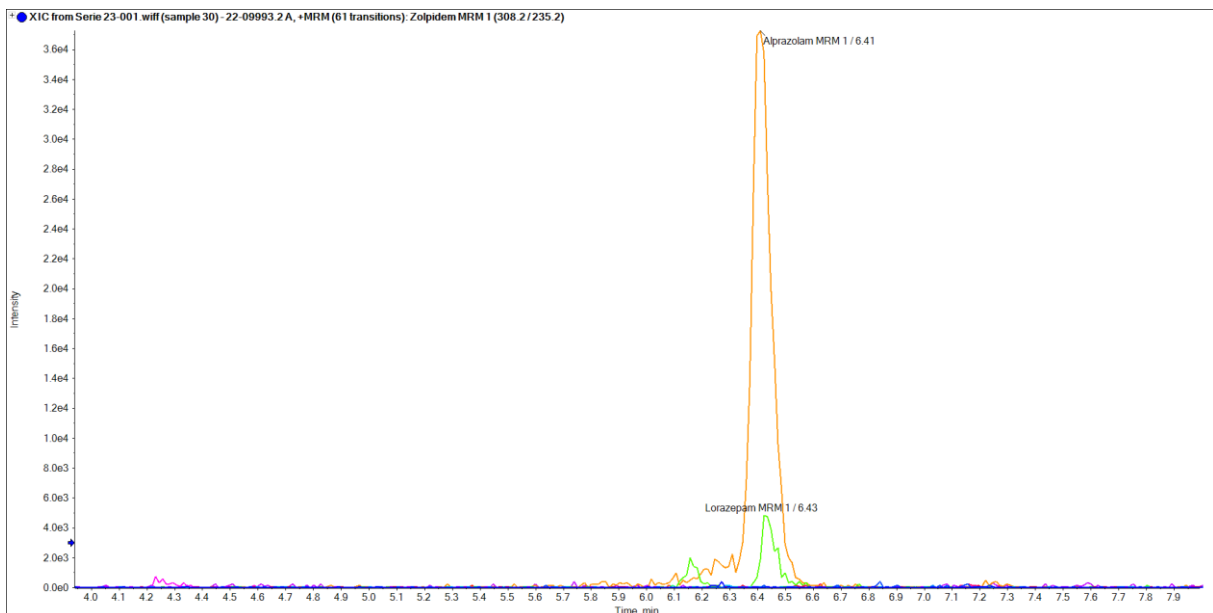


Blood sample 34

ReproSil Star Phenyl-Ether - Dr. Maisch HPLC GmbH

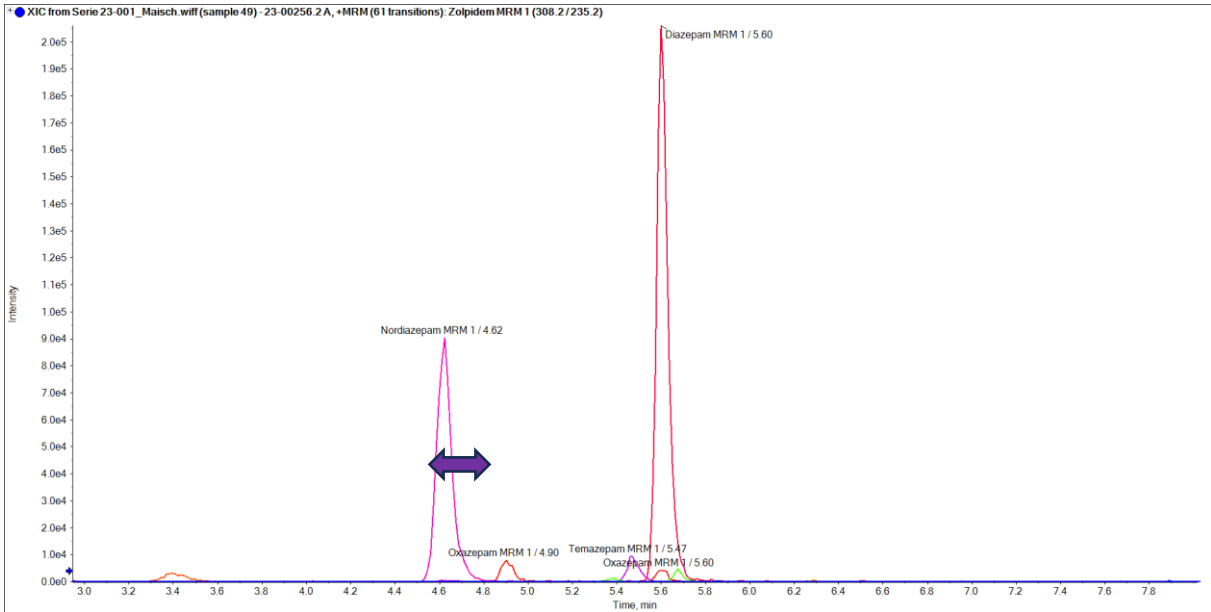


Synergi Polar RP - Phenomenex

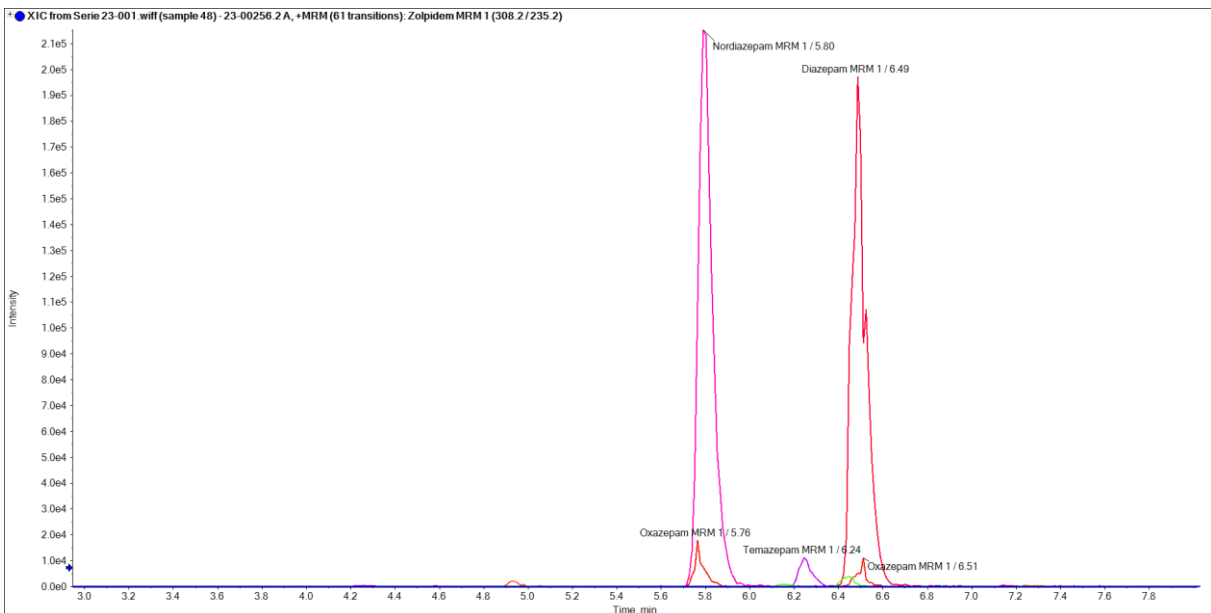


Blood sample 49

ReproSil Star Phenyl-Ether - Dr. Maisch HPLC GmbH



Synergi Polar RP - Phenomenex



Application courtesy of Dr. Stefan König, Institut für Rechtsmedizin Bern (Switzerland), Forensische Toxikologie und Chemie.

Dr. Maisch products are available globally. For more information contact your Dr. Maisch representative at info@dr-maisch.com.

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