

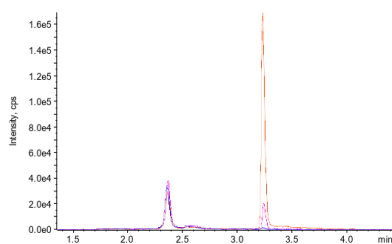
## Standards and IS, Clarity OTX Central Nervous System (CNS) Tissue

**Column:** bioZen™ 2.6 µm Oligo,LC Column 100 x 2.1 mm, Ea  
**Dimensions:** 100 x 2.1 mm ID  
**Order No:** 00D-4790-AN  
**Elution Type:** Gradient  
**Eluent A:** 1.0 % HFIP, 0.1% DIPEA in Water with 10 µm EDTA  
**Eluent B:** 0.075% HFIP, 0.0375% DIPEA in Water:ACN (35:65) with 10 µm EDTA



Gradient Profile:	Step No.	Time (min)	Pct A	Pct B
	1	0	95	5
	10	6.2	80	20
	11	6.3	20	80
	12	7.5	5	95
	13	7.8	5	95
	14	7.9	95	5
	2	0.1	95	5
	3	2.5	80	20
	4	4	40	60
	5	4.1	5	95
	6	4.4	5	95
	7	5	80	20
	8	5.8	5	95
	9	6.1	5	95

**Flow Rate:** 0.5 mL/min  
**Col. Temp.:** 80 °C  
**Detection:** LC/MS/MS @ (ambient)



## Standards and IS, Clarity OTX Central Nervous System (CNS) Tissue

**ANALYTES:**



# Sample Preparation Details

for HPLC Application ID No.: 26262

## Standards and IS, Clarity OTX Central Nervous System (CNS) Tissue

### PRODUCT DESCRIPTION:

Clarity® OTX, 100 mg / well, 96-Well Plates , Ea

Order No.: 8E-S103-EGA

### 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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Aliquot 75 µL calibration standards, QC samples, study samples onto 2 mL 96-well round bottom plate  
Add 30 µL IS (1 µg/mL) to all wells except blank controls (add 30 µL water to blanks)  
Condition 96-well plate with 1 mL MeOH  
Equilibrate with 1 mL Equilibration Buffer (50 mM Ammonium Acetate, pH 5.5)

#### Wash:

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#### Dry:

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#### Elute:

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#### Final Prep and Analysis:

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Inject: 2 µL on HPLC LC/MS/MS @ (ambient)

ANALYTES:	Spiked Conc. (ng/mL)	Log P	pKa	% Rec	%RSC (n=0)
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**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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