



**FENTANYL / NORFENTANYL ON ORAL SWABS USING:  
200 mg CLEAN SCREEN<sup>®</sup> EXTRACTION COLUMN**  
PART #: ZSDAU020  
LC-MSMS

**1. PREPARE SAMPLE**

**Preparation of Standards:**

To separate tube add 0, 1, 5, 10 ng of Fentanyl / Norfentanyl in methanol. Evaporate off the solvent. Add 100  $\mu$ L of drug free oral fluid. Vortex mix and allow to stand for 30 minutes. Take clean, dry (drug free) swab and swab up the oral fluid and allow standing for 15 minutes. Remove oral swab.

**SAMPLE PRE TREATMENT:**

To 200  $\mu$ L of methanol (pH 6) add internal standard.\*  
Insert oral swab into methanol and mix for 1 minute, add a further 100  $\mu$ L of methanol, allow to stand for 10 minutes. Remove swab and 3 mL of 100 mM phosphate buffer (pH 6). Vortex and centrifuge as appropriate.

**2. CONDITION COLUMN:**

1 x 3 mL MeOH

1 x 3 mL H<sub>2</sub>O

1 x 1 mL 100 mM phosphate buffer (pH 6)

**Note:** aspirate at < 3 inches Hg to prevent sorbent drying out

**3. APPLY SAMPLE:**

Load sample at 1-2 mL / minute.

**4. WASH COLUMN:**

1 x 3 mL DI Water

1 x 3 mL 1% acetate acid

1 x 3 mL Methanol

Dry column (5 minutes at > 10 inches Hg).

**5. ELUTE FENTANYL/ NORFENTANYL:**

1 x 3 mL ethyl acetate; acetonitrile: ammonia (78: 20: 2 v/v)

Collect eluate at 1-2 mL /minute.

**6. EVAPORATION:**

Evaporate eluates under a gentle stream of nitrogen < 40°C.

**7. RECONSTITUTE sample in 20  $\mu$ L of methanol.**

Inject 5  $\mu$ L.

## INSTRUMENT CONDITIONS:

**Column:** 150 x 2.1 mm (3 µm) PFP Thermofisher

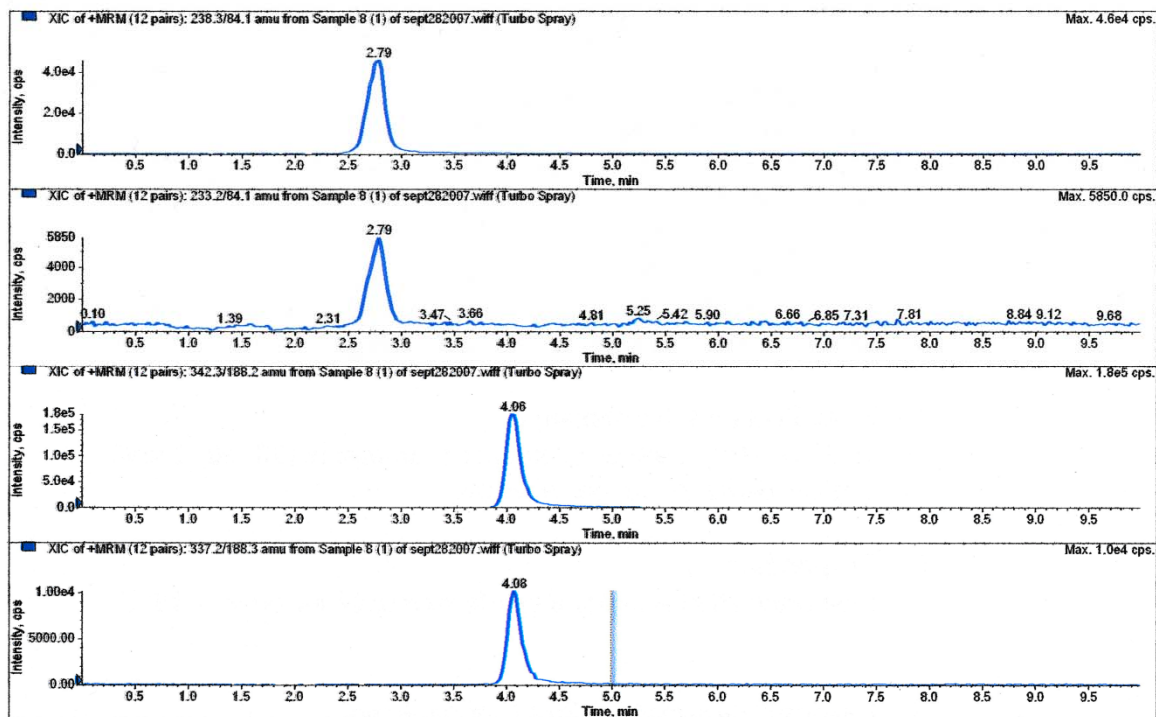
**Mobile phase:** Acetonitrile: 0.1% Formic acid (60:40)

**Flowrate:** 0.35 mL/minute

**Column Temperature:** ambient

**Detector:** API 2000 MS/MS

<u>Compound</u>	<u>MRM Transition</u>	<u>Cerilliant #</u>
Fentanyl	333.2/188.3	F-002
*Fentanyl-D5	342.2/188.2	F-001
Norfentanyl	233.2/84.1	N-031
*Norfentanyl-D5	238.3/84.1	N-030



**Recovery:** > 90% (N=100)

**LOD:** 1 ng/mL