510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K241969

B Applicant

Qingdao HIGHTOP Biotech Co., Ltd.

C Proprietary and Established Names

Hightop® Home Use Fentanyl/Norfentanyl Urine Rapid Test Panel; Hightop® Fentanyl/Norfentanyl Urine Rapid Test Panel

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
NGL	Class II	21 CFR 862.3650 - Opiate Test System	TX - Clinical Toxicology

II Submission/Device Overview:

A Purpose for Submission:

New Device

B Measurand:

Fentanyl and Norfentanyl

C Type of Test:

lateral flow immunochromatographic assay

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

Hightop® Home Use Fentanyl/Norfentanyl Urine Rapid Test Panel is a competitive binding, lateral flow immunochromatographic assay for qualitative detection of fentanyl and norfentanyl, the major metabolite of fentanyl in human urine at the cut-off concentrations listed below:

Analyte	Cut-off Level
Fentanyl (FYL)	1ng/mL
Norfentanyl (NFYL)	5ng/mL

The test is available in a single test of FYL or NFYL or a Double panel of FYL and NFYL. It is intended for OTC use.

The test provides only a preliminary test result. A more specific alternative chemical method must be used in order to obtain a confirmed analytical result. GC/MS or LC/MS is the preferred confirmatory method.

Hightop® Fentanyl/Norfentanyl Urine Rapid Test Panel is a competitive binding, lateral flow immunochromatographic assay for qualitative detection of fentanyl and norfentanyl, the major metabolite of fentanyl in human urine at the cut-off concentrations listed below:

Analyte	Calibrator	Cut-off Level
Fentanyl (FYL)	Fentanyl	1ng/mL
Norfentanyl (NFYL)	Norfentanyl	5ng/mL

The test is available in a single test of FYL or NFYL or a Double panel of FYL and NFYL.

The test panel provides only a preliminary test result. A more specific alternative chemical method must be used in order to obtain a confirmed analytical result. GC/MS or LC/MS is the preferred confirmatory method.

The test panel is not intended to distinguish between prescription use or abuse of fentanyl. Clinical consideration and professional judgment should be applied to the test result, particularly in evaluating a preliminary positive result.

C Special Conditions for Use Statement(s):

OTC - Over The Counter

D Special Instrument Requirements:

Not applicable

IV Device/System Characteristics:

A Device Description:

The Hightop® Home Use Fentanyl/Norfentanyl Urine Rapid Test Panel and Hightop® Fentanyl/Norfentanyl Urine Rapid Test Panel are immunoassays intended for the qualitative detection of fentanyl and norfentanyl in human urine. Each Hightop® fentanyl/norfentanyl urine test device consists of a Test Panel and a package insert. Each Test Panel is sealed with sachets of desiccant in an aluminum pouch.

B Principle of Operation:

These devices are rapid lateral flow immunoassays. To perform the test, the user will pull off the cap on the device and activate the test by dipping the sample pads into human urine sample for 5-10 seconds. Urine sample will be absorbed by the sample pad and move up to the conjugate pad that contains anti-drug antibody colloidal gold conjugates. The urine sample will continue to move up to the nitrocellulose membrane where drug protein is coated onto the Test Region. If there is no drug present or the drug concentration in the specimen is below cutoff level, the red colloidal gold conjugate will bind to the drug conjugate at the Test Region, to form a visible band which indicated a negative result. If there is drug present in the specimen at above cutoff level, the drug will bind to the limited antibodies on colloidal gold, leaving no antibody available for binding to the drug conjugates on membrane. Thus, the absences of a test line band present at Test Region indicates a presumptive positive result for that drug.

The Control Region is to indicate that the test has performed properly and should appear regardless of the presence of the drug. This is a built-in internal control of the device. If control line does not appear, the test result is invalid.

V Substantial Equivalence Information:

A Predicate Device Name(s):

AllTest Fentanyl Urine Test Cassette

B Predicate 510(k) Number(s):

K233417

C Comparison with Predicate(s):

Device & Predicate Device(s):	K241969	K233417
Device Trade Name	Hightop® Home Use Fentanyl/Norfentanyl Urine Rapid Test Panel Hightop® Fentanyl/Norfentanyl Urine Rapid Test Panel	AllTest Fentanyl Urine Test Cassette
General Device		

Characteristic Similarities		
Intended Use/Indications For Use	For the qualitative determination of fentanyl in human urine.	Same
Methodology	Competitive binding, lateral flow immunochromatographic assays based on the principle of antigen antibody immunochemistry.	Same
Specimen Type	Urine	Same
General Device Characteristic Differences		
Calibrator and Cut-Off Values	Fentanyl (FTY) 1 ng/ml Norfentanyl (NFTY) 5 ng/ml	Fentanyl (FTY) 1 ng/ml
Configuration	Panel	Cassette

VI Standards/Guidance Documents Referenced:

Not applicable.

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

Precision studies were carried out for samples with concentrations of -100% cut off, -75% cut off, -50% cut off, -25% cut off, cut off, +25% cut off, +50% cut off, +75% cut off and +100% cut off for each target drug. These samples were prepared by spiking fentanyl or norfentanyl in negative urine samples. Each fentanyl or norfentanyl concentration was confirmed by LC/MS. All sample aliquots were blindly labeled by the person who prepared the samples and didn't take part in the sample testing. Three operators tested blinded urine samples over a 10-day period in randomized order with six tests per day per concentration per lot per operator (6 replicate per day per lot x 9 concentrations) for a total of 54 tests per day per operator.

Fentanyl

Lot Number	-100% cut off	-75% cut off	-50% cut off	-25% cutoff	cut off	+25% cut off	+50% cut off	+75% cut off	+100% cut off
Lot 1	60-/0+	60-/0+	60-/0+	60-/0+	40+/20-	60+/0-	60+/0-	60+/0-	60+/0-
Lot 2	60-/0+	60-/0+	60-/0+	59-/1+	38+/22-	60+/0-	60+/0-	60+/0-	60+/0-
Lot 3	60-/0+	60-/0+	60-/0+	59-/1+	38+/22-	60+/0-	60+/0-	60+/0-	60+/0-

Norfentanyl

Lot Number	-100% cut off	-75% cut off	-50% cut off	-25% cutoff	cut off	+25% cut off	+50% cut off	+75% cut off	+100% cut off
Lot 1	60-/0+	60-/0+	60-/0+	58-/2+	35+/25-	60+/0-	60+/0-	60+/0-	60+/0-
Lot 2	60-/0+	60-/0+	60-/0+	58-/2+	32+/28-	60+/0-	60+/0-	60+/0-	60+/0-
Lot 3	60-/0+	60-/0+	60-/0+	59-/1+	33+/27-	60+/0-	60+/0-	60+/0-	60+/0-

2. Linearity:

Not applicable. These devices are intended for qualitative use only.

3. Analytical Specificity/Interference:

Potential interfering substances found in human urine of physiological or pathological conditions were added to drug-free urine and target drug fentanyl urine with concentrations at 50% below and 50% above Cut-Off levels. These urine samples were tested using three batches of each device. Compounds that showed no interference at a concentration of 100µg/mL or specified concentrations are summarized in the following tables.

Acetaminophen	Ecgonine methyl ester	Oxolinic acid
Acetone (1000 mg/dL)	Ephedrine	Oxymetazoline
Acetophenetidin	Erythromycin	Papaverine
Acetylsalicylic acid	Estradiol	Penicillin G
Albumin (100 mg/dL)	Estrone	Perphenazine
Albuterol	Ethanol (1%)	Phencyclidine
7-Aminonitrazepam	Fenfluramine	Phenelzine
Amitriptyline	Fenofibrate	Phenobarbital
Amlodipine besylate	Fenoprofen	Phentermine
Amobarbital	Fluphenazine	Phenylethylamine
Amoxicillin	Fotemustine	Prednisone
Ampicillin	Furosemide	Promazine
Apomorphine	Galactose	Promethazine
Ascorbic acid	γ-Globulin (500 mg/dL)	Propoxyphene
Aspartame	Gemfibrozil	Propranolol
Aspirin	Gentisic acid	Pseudoephedrine
Atropine	Glucose (3000 mg/dL)	Pyridoxine

Baclofen	Guaiacolglyceryl ether	Pyrilamine
Benzilic acid	Hemoglobin	Pyrogallol
Benzocaine	Hexobarbital	Quinidine
Benzoic acid	Hydralazine	Quinine
Benzoylecgonine	Hydrochlorothiazide	Quinolinic Acid
Benzylpiperiazine	Hydrocortisone	Ranitidine
Bilirubin	Hydroxybutyric Acid	Riboflavin
Boric Acid (1%)	Ibuprofen	Salicylic acid
Bromo-2,5,Dimethoxyphenethylamine	Imipramine	Secobarbital
Bupropion	Isoproterenol	Serotonin
Caffeine	Isoxsuprine (10 µg/mL)	Sodium Azide
Carbamazepine	Ketoprofen	Sulfamethazine
Carisoprodol	Labetalol	Sulindac
Cetirizine	Lamotrigine	Tetracycline
Chloral hydrate	Lidocaine	Tetrahydrocortisone3-(β-Dglucuronide)
Chloramphenicol	Lisinopril	Tetrahydrocortisone 3-acetate
Chlordiazepoxide	Loperamide	Tetrahydrozoline
Chlorothiazide	Loratidine	Thiamine
Chlorpheniramine	Maprotiline	Triamterene
Chlorpromazine	Meperidine	Trifluoperazine
Cholesterol	Meprobamate	Trifluoromethylphenyl-piperazine
Clofibrate	Methapyrilene	Trimethoprim
Clomipramine	Methaqualone	Tryptamine
Clonidine	Methoxyphenamine	Tyramine
Cortisone	Methylphenidate	Urea (2000 mg/dL)
Cotinine	Metoprolol	Uric acid
Creatine Hydrate	Metronidazole	Valproic acid (250 µg/mL)
Creatinine	N-Acetylprocainamide	Venlafaxine
Cyclobenzaprine	N-desmethyl Tapentadol	Verapamil
γ-Cyclodextrin	Nacl (4000 mg/dL)	Zolpidem
Cyproheptadine	Nalidixic acid	Zomepirac
Demoxepam	Naproxen	7-Aminoflunitrazepam
Deoxycorticosterone	Niacinamide	Metformin
Desipramine	Nicotine	Norpseudoephedrine
Diclofenac	Nicotinic Acid	Oxazepam Glucuronide
Diflunisal	Nifedipine	Lorazepam Glucuronide
Digoxin	Norethindrone	LSD
Dimethyl-aminoantipyrine	Norpropoxyphene	THC
Diphenhydramine	Nortriptyline	L-thyroxine
Diphenylhydantoin	Noscapine	Dextromethorphan
DL-Tryptophan	O-Hydroxyhippuric acid	Ketamine
DL-Tyrosine	Octopamine	Thioridazine

Dopamine (Hydroxytyramine)	Oxalic acid (100mg/dL)	
Doxepin	Oxazepam	

Specificity

To test specificity, drug metabolites and other components that are likely to interfere in urine samples were tested using three batches of the device. The lowest concentration that caused a positive result for each compound are listed below. If no cross reactivity was observed the highest concentration tested is shown.

Fentanyl (Cutoff=1ng/mL)	**Minimum concentration required to obtain a positive result (ng/mL)**	**% Cross-Reactivity**
Fentanyl	1	100
Norfentanyl	>100000	<0.001
4-Fluoro-isobutyryl fentanyl	200	0.5
9-Hydroxy Risperidone	10000	0.01
Acetyl fentanyl	5	20
Acetyl Norfentanyl	>100000	<0.001
(±)-β-hydroxythiofentanyl	25	4
Acryl fentanyl	5	20
Alfentanil	10000	0.01
Butyryl fentanyl	10	10
Carfentanil	10	10
(±)-cis-3-methylfentanyl	1000	0.1
Despropionyl fentanyl (4-ANPP)	10000	0.01
Furanyl Fentanyl	20	5
Isobutyryl Fentanyl	50	2
Labetalol Hydrochloride	>100000	<0.001
MT-45	10000	0.01
Norcarfentanil	>20000	<0.005
Ocfentanil	1000	0.1
Para-fluorobutyryl Fentanyl (PFBF)	20	5
Para-fluoro Fentanyl	20	5
Remifentanil	>20000	<0.005
Risperidone	1000	0.1
Sufentanil	1000	0.1
Thienyl Fentnayl	1000	0.1
Trans-d, I 3-Methylfentanyl	1000	0.1
Trazodone	1000	0.1
U-47700	>100000	<0.001

Fentanyl (Cutoff=1ng/mL)	Minimum concentration required to obtain a positive result (ng/mL)	% Cross-Reactivity
Valeryl fentanyl	50	2
ω-1-Hydroxyfentanyl	>20000	<0.005

Norfentanyl (Cutoff=5ng/mL)	Minimum concentration required to obtain a positive result	%Cross-Reactivity
Norfentanyl	5	100
Fentanyl	9	55.6
4-Fluoro-isobutyryl fentanyl	>20,000	<0.025
9-Hydroxy Risperidone	10,000	0.05
Acetyl fentanyl	150	3.3
Acetyl Norfentanyl	200	2.5
(±)-β-hydroxythiofentanyl	20	25
Acryl fentanyl	50	10
Alfentanil	1,000	0.5
Butyryl fentanyl	10	50
Carfentanil	10,000	0.05
(±)-cis-3-methylfentanyl	50	10
Despropionyl fentanyl (4-ANPP)	>20,000	<0.025
Furanyl Fentanyl	50	10
Isobutyryl Fentanyl	1,000	0.5
Labetalol Hydrochloride	>100,000	<0.005
MT-45	5,000	0.1
Norcarfentanil	>20,000	<0.025
Ocfentanil	500	1
Para-fluorobutyryl Fentanyl (PFBF)	20	25
Para-fluoro Fentanyl	10	50
Remifentanil	10,000	0.05
Risperidone	1,000	0.5
Sufentanil	1000	0.5
Thienyl Fentnayl	50	10
Trans-d, I 3-Methylfentanyl	50	10
Trazodone	10,000	0.05
U-47700	>100,000	<0.005
Valeryl fentanyl	>20,000	<0.025
ω-1-Hydroxyfentanyl	>20,000	<0.025

The following opioids compounds were tested at a concentration of 100ug/mL. Negative results were obtained for all these compounds. There is no cross-reactivity for these compounds using the Hightop® Device.

6-Acetyl morphine	Naloxone
Amphetamine	Naltrexone
Buprenorphine	Norbuprenorphine
Buprenorphineglucuronide	Norcodeine
Codeine	Norketamine
Dextromethorphan	Normeperidine
Dihydrocodeine	Normorphine
EDDP	Noroxycodone
EMDP	Oxycodone
Fluoxetine	Oxymorphone
Heroin	Pentazocine (Talwin)
Hydrocodone	Pipamperone
Hydromorphone	Tapentadol
Ketamine	Thioridazine
Levorphanol	Tilidine
Meperidine	Tramadol
Methadone	Tramadol-O-Desmethyl
Morphine	Tramadol-N-Desmethyl
Morphine-3-glucuronide	

Effect of Urine Specific Gravity and Urine pH

To investigate the effect of urine specific gravity and urine pH, urine samples, with 1.000, 1.003, 1.006, 1.009, 1.011, 1.014, 1.017, 1.02, 1.023, 1.027, 1.031, and 1.035 specific gravity or urine samples with pH 4, 5, 6, 7, 8,and 9 were spiked with targets fentanyl and norfentanyl at 50% below and 50% above Cut-Off levels. These samples were tested using three lots of device. Results were all positive for samples at and above +50% Cut- Offs and all negative for samples at and below -50% Cut-Offs.

Read Time Study

Fentanyl and Norfentanyl Standards with concentrations of -50% cutoff, +50% cutoff, and drug free urine were tested using three lots of the device and read at 10 time points (i.e. 3rd, 4th, 5th, 10th, 15th, 20th, 25th, 30th, 35th, 40th minutes) by three different operators according to procedures in the product insert. All the results were consistent with the required reading time at 5 minutes and not more than 10 minutes.

4. Assay Reportable Range:

Characterization of how the device performs analytically around the claimed cutoff concentration appears in the precision section VII.A.1 above.

5. Traceability, Stability, Expected Values (Controls, Calibrators, or Methods):

All drug calibrators of the device are traceable to available commercial reference materials.

6. Detection Limit:

Characterization of how the device performs at low concentrations appears in the precision section VII.A.1 above.

7. Assay Cut-Off:

Characterization of how the device performs analytically around the claimed cutoff concentration appears in the precision section VII.A.1 above.

B Comparison Studies:

1. Method Comparison with Predicate Device:

Method comparison studies for 1 lot of the Hightop® Fentanyl/Norfentanyl Urine Rapid Test Panel were performed by three different operators. Operators ran 80 (40 negative and 40 positive) unaltered clinical samples. The samples were blind labeled and compared to LC/MS results. The results are presented in the tables below.

Fentanyl

		Negative	Low Negative by LC/MS (less than -50%)	Near Cutoff Negative by LC/MS (Between -50% and cutoff)	Near Cutoff Positive by LC/MS (Between the cutoff and +50%)	High Positive by LC/MS (greater than +50%)
Operator 1	Positive	0	0	2	19	20
	Negative	10	13	15	1	0
Operator 2	Positive	0	0	2	19	20
	Negative	10	13	15	1	0
Operator 3	Positive	0	0	1	19	20
	Negative	10	13	16	1	0

Discordant Results

Operator	Sample ID	LC/MS Result (ng/mL)	Rapid Test Result
Operator 1, 3	SE086 & SE156	0.7880	Positive

Operator	Sample ID	LC/MS Result (ng/mL)	Rapid Test Result
Operator 1, 2	SE052 &SE169	0.7912	Positive
Operator 1	SE031	1.0078	Negative
Operator 2	SE110	0.7874	Positive
Operator 2	SE182	1.0282	Negative
Operator 3	SE240	1.0811	Negative

Norfentanyl

		Negative	Low Negative by LC/MS (less than -50%)	Near Cutoff Negative by LC/MS (Between -50% and cutoff)	Near Cutoff Positive by LC/MS (Between the cutoff and +50%)	High Positive by LC/MS (greater than +50%)
Operator 1	Positive	0	0	1	19	20
	Negative	10	13	16	1	0
Operator 2	Positive	0	0	2	18	20
	Negative	10	13	15	2	0
Operator 3	Positive	0	0	1	19	20
	Negative	10	13	16	1	0

Discordant Results

Operator	Sample ID	LC/MS Result (ng/mL)	Rapid Test Result
Operator 1, 2	SF133 & SF107	4.6743	Positive
Operator 1	SF016	5.1608	Negative
Operator 2, 3	SF201 & SF219	4.1946	Positive
Operator 2	SF231	5.1303	Negative
Operator 2, 3	SF088 & SF221	5.0744	Negative

2. Matrix Comparison:

Not applicable. These devices are for use with urine samples only.

C Clinical Studies:

1. Clinical Sensitivity:

Not applicable.

2. Clinical Specificity:

Not applicable.

3. Other Clinical Supportive Data (When 1. and 2. Are Not Applicable):

A lay user study was performed at three testing sites representative of intended use sites with 140 lay users. The lay users had diverse educational and professional backgrounds and ranged in age from 18 to >50 years. Three lots of the device were used in the study. Urine samples were prepared at the following concentrations; -100%, +/-75%, +/-50%, +/-25% of the cut-offs by spiking drug(s) into drug free-pooled urine specimens. The concentrations of the samples were confirmed by LC/MS. Each sample was aliquoted into individual containers, blind-labeled and randomized. Each participant was provided with the package insert, 1 blind labeled sample and a device. The results are summarized below:

% of Cutoff	Number of samples	Fentanyl Concentration by LC/MS (ng/mL)	Lay person results		The percentage of correct results (%)
			No. of Positive	No. of Negative	
-100% Cutoff	20	0	0	20	100.0%
-75% Cutoff	20	0.24	0	20	100.0%
-50% Cutoff	20	0.47	0	20	100.0%
-25% Cutoff	20	0.72	0	20	100.0%
+25% Cutoff	20	1.21	20	0	100.0%
+50% Cutoff	20	1.50	20	0	100.0%
+75% Cutoff	20	1.81	20	0	100.0%

% of Cutoff	Number of samples	Norfentanyl Concentration by LC/MS (ng/mL)	Lay person results		The percentage of correct results (%)
			No. of Positive	No. of Negative	
-100% Cutoff	20	0	0	20	100.0%
-75% Cutoff	20	1.33	0	20	100.0%
-50% Cutoff	20	2.63	0	20	100.0%
-25% Cutoff	20	4.06	1	19	95.0%
+25% Cutoff	20	6.03	20	0	100.0%
+50% Cutoff	20	7.29	20	0	100.0%
+75% Cutoff	20	9.08	20	0	100.0%

Lay-users were also given surveys on the ease of understanding the package insert instructions. All lay users indicated that the device instructions can be easily followed. A

Flesch-Kincaid reading analysis was performed on the package insert and the score revealed a reading grade level of less than 7.

D Clinical Cut-Off:

Not applicable.

E Expected Values/Reference Range:

Not applicable.

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

IX Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.