510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K231007

B Applicant

Microgenics Corporation

C Proprietary and Established Names

CEDIA™ Heroin Metabolite (6-AM) Assay

D Regulatory Information

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

II Submission/Device Overview:

A Purpose for Submission: New device.

B Measurand:

6-Acetylmorphine (6-AM).

C Type of Test:

Qualitative and Semi-quantitative Homogeneous Enzyme Immunoassay

III Intended Use/Indications for Use:

A Intended Use(s):

The CEDIA™ Heroin Metabolite (6-AM) Assay is a homogeneous enzyme immunoassay for the in vitro qualitative and/or semi-quantitative determination of the presence of heroin metabolite (6-AM) in human urine at a cut-off concentration of 10 ng/mL. The assay is intended to be used in

laboratories and provides a rapid analytical screening procedure to detect 6-Acetylmorphine in human urine. The assay is designed for use with a number of clinical chemistry analyzers. This product is intended to be used by trained professionals only.

The semi-quantitative mode is for the purpose of enabling laboratories to determine an appropriate dilution of the specimen for confirmation by a confirmatory method such as Liquid Chromatography/tandem mass spectrometry (LC-MS/MS) or permitting laboratories to establish quality control procedures. The assay provides only a preliminary analytical test result. A more specific alternative chemical method must be used to obtain a confirmed analytical result. Gas chromatography/mass spectrometry (GC/MS) or Liquid chromatography/ mass spectrometry (LC-MS/MS) is the preferred confirmatory method.

Clinical and professional judgment should be applied to any drug of abuse test result, particularly when preliminary results are used. For In Vitro Diagnostic Use Only.

B Indication(s) for Use:
See Intended use above

C Special Conditions for Use Statement(s):
Rx - For Prescription Use Only

D Special Instrument Requirements:

Performance characteristics studies were conducted on the Horiba Yumizen C1200 clinical chemistry analyzer.

IV Device/System Characteristics:

A Device Description:

CEDIA™ Heroin Metabolite (6-AM) assay is supplied as a two liquid (1 and 2) and two lyophilized reagents (1a and 2a) kit that is available in a kit configuration.

1. EA Reconstitution Buffer: Contains 0.32 mg/L mouse monoclonal antibodies to 6-Acetylmorphine, buffer salts, stabilizer, detergent and preservative

1a. EA Reagent: Contains 0.171 g/L Enzyme Acceptor, buffer salts, detergent and preservative

2. ED Reconstitution Buffer: Contains buffer salts, stabilizer, and preservative

2a. ED Reagent: Contains 16.2 µg/L Enzyme Donor conjugated to 6- Acetylmorphine, 1.67 g/L chlorophenol red-β-D galactopyranoside, stabilizer, detergent, preservative.

B Principle of Operation: CEDIA technology uses recombinant DNA technology to produce a unique homogeneous enzyme immunoassay system. The assay is based on the bacterial enzyme β-galactosidase, which has been genetically engineered into two inactive fragments. These

fragments spontaneously re-associate to form fully active enzymes that, in the assay format, cleave a substrate. This generates a color change that can be measured spectrophotometrically.

In the CEDIA™ Heroin Metabolite (6-AM), the analyte in the sample competes with 6-AM conjugated to Enzyme Donor (ED) for antibody binding sites. If 6-AM is present in the sample, it binds to antibody, leaving the ED-6-AM conjugate free to re-associate with Enzyme Acceptor (EA) to form active β-galactosidase. If no 6-AM is present in the sample, antibody binds to the ED-6-AM conjugate, inhibiting the re-association of inactive - galactosidase fragments, and thus reducing the amount of active enzyme formed. The amount of active enzyme formed, and resultant absorbance change are proportional to the amount of 6-AM present in the sample.

V Substantial Equivalence Information:

A Predicate Device Name(s):
CEDIA™ Heroin Metabolite (6-AM) assay

B Predicate 510(k) Number(s):
K192943

C Comparison with Predicate(s):

Device & Predicate Device(s):	K231007	K192943
Device Trade Name	CEDIA™ Heroin Metabolite (6-AM) assay	CEDIA™ Heroin Metabolite (6-AM) assay
General Device Characteristic Similarities		
Intended Use/Indications For Use	Homogeneous enzyme immunoassay for the in vitro qualitative and/or semi-quantitative determination of the presence of heroin metabolite (6-AM) in human urine at a cut-off concentration of 10 ng/mL.	Same
Analyte	6-Acetylmorphine (6-AM)	6-Acetylmorphine (6-AM)
General Device Characteristic Differences		
Instrument	Horiba Yumizen C1200	Horiba Pentra C400

VI Standards/Guidance Documents Referenced:

CLSI EP05-A3 – Evaluation of Precision of Quantitative Measurement Procedures; Approved Guideline – Third Edition.

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

All analytical performance studies were conducted on the Horiba Yumizen C1200 clinical chemistry analyzer.

1. Precision/Reproducibility:

The precision study was performed based upon recommendations in CLSI Guideline EP05-A3, at one site with one analyzer and two lots of reagents. Testing was carried out for 20 days with two runs per day, at least two hours apart and two replicates per run in both qualitative and semi-quantitative modes, giving a total of 80 determinants (n = 80). Drug-free negative urine was spiked with 6- Acetylmorphine analyte to final concentrations of -100%, -75%, -50%, -25%, below cutoff, cutoff, and +25%, +50%, +75% and +100%, above cutoff, and the concentrations were confirmed by LC-MS/MS or GC/MS.

For qualitative and semi-quantitative mode results from a representative lot are summarized in the tables below.

Precision Data in Qualitative Mode

% of Cutoff	Spiked Conc. (ng/mL)	# of Determinants	# Negative/ # Positive
-100	0	80	80 / 0
-75	2.5	80	80 / 0
-50	5	80	80 / 0
-25	7.5	80	80 / 0
100	10	80	36 / 44
+25	12.5	80	0 / 80
+50	15	80	0 / 80
+75	17.5	80	0 / 80
+100	20	80	0 / 80

Precision Data in Semi-Quantitative Mode

% of Cutoff	Spiked Conc. (ng/mL)	# of Determinants	# Negative/ # Positive	Within-run CV (%)	Total-run CV (%)
-100	0	80	80 / 0	N/A	N/A

-75	2.5	80	80 / 0	9.5	14.5
-50	5	80	80 / 0	3.9	6.2
-25	7.5	80	80 / 0	3.8	5.8
100	10	80	31 / 49	3.6	5.4
+25	12.5	80	0 / 80	3.0	4.9
+50	15	80	0 / 80	3.4	4.9
+75	17.5	80	0 / 80	2.7	3.7
+100	20	80	0 / 80	1.9	3.3

2. Linearity:

To demonstrate linearity of the assay throughout the calibration range of 0 to 20 ng/mL, a drug free–urine pool spiked with 6-AM at 20 ng/mL was serially diluted with drug free urine to generate ten intermediate levels. Each sample was run in replicates of five in semi-quantitative mode and the average was used to determine percent recovery compared to the expected target value.

The observed result (y) and the target expected result (x) were compared using the least squares regression method. The regression equation and correlation obtained are:

$$y = 0.9008x + 0.3445; R^2 = 0.997$$

The recovery of the samples from the linear range of the assay (2 ng/mL to 20 ng/mL) prepared by dilution ranged from 90.5% to 111.0%.

3. Analytical Specificity/Interference:

A. Cross-reactivity (specificity) of the structurally related and unrelated compounds with the performance of the candidate device was evaluated by adding known amounts of each compound to drug-free negative urine. The samples were tested in duplicate with the candidate device in both the qualitative and semi-quantitative modes. Percent cross-reactivity was calculated as follows:

% Cross-reactivity = (Cutoff concentration / Lowest concentration of cross reactant that gives a positive result) x 100

Cross-Reactivity of 6-Acetylmorphine and Heroin:

6-Acetylmorphine and Heroin	Tested Concentration (ng/mL)	Assay Result	Cross-Reactivity (%)
6-Acetylmorphine	10	Pos	100%
Heroin	300	Pos	3%

Cross-Reactivity of Opiates and Structurally Related Compounds:

Structurally related compounds and other opiates	Tested Concentration (ng/mL)	Assay Result	Cross-Reactivity (%)
6-Acetylcodeine	50,000	Pos	0.02%
Buprenorphine	100,000	Neg	No Cross-Reactivity
Buprenorphine-3β-D-glucuronide	100,000	Neg	No Cross-Reactivity
Codeine	100,000	Neg	No Cross-Reactivity
Dextromethorphan	90,000	Pos	0.01%
Dihydrocodeine	100,000	Neg	No Cross-Reactivity
EDDP	100,000	Neg	No Cross-Reactivity
EMDP	100,000	Neg	No Cross-Reactivity
Ethylmorphine	100,000	Neg	No Cross-Reactivity
Fentanyl	100,000	Neg	No Cross-Reactivity
Hydrocodone	100,000	Neg	No Cross-Reactivity
Hydromorphone	20,000	Pos	0.04%
Hydromorphone-3β-D-glucuronide	100,000	Neg	No Cross-Reactivity
Levorphanol	15,000	Pos	0.05%
Methadone	100,000	Neg	No Cross-Reactivity
Meperidine	100,000	Neg	No Cross-Reactivity
Mitragynine	100,000	Neg	No Cross-Reactivity
7-Hydroxymitragynine	100,000	Neg	No Cross-Reactivity
Morphine	13,500	Pos	0.07%
Morphine-3β-D-Glucuronide	100,000	Neg	No Cross-Reactivity
Morphine-6β-D-Glucuronide	100,000	Neg	No Cross-Reactivity
Nalorphine	10,500	Pos	0.10%
Naloxone	100,000	Neg	No Cross-Reactivity
Naltrexone	100,000	Neg	No Cross-Reactivity
Norbuprenorphine	100,000	Neg	No Cross-Reactivity
Norbuprenorphine Glucuronide	100,000	Neg	No Cross-Reactivity
Norcodeine	100,000	Neg	No Cross-Reactivity
Norhydrocodone	100,000	Neg	No Cross-Reactivity
Normorphine	50,000	Pos	0.02%
Norpropoxyphene	100,000	Neg	No Cross-Reactivity
Noroxycodone	100,000	Neg	No Cross-Reactivity
Noroxymorphone	100,000	Neg	No Cross-Reactivity
Oxymorphone-3β-D- glucuronide	100,000	Neg	No Cross-Reactivity
Oxycodone	100,000	Neg	No Cross-Reactivity

Oxymorphone	100,000	Neg	No Cross-Reactivity
Tapentadol	100,000	Neg	No Cross-Reactivity
Tramadol	100,000	Neg	No Cross-Reactivity

B. Interference: The interference study was performed using one lot of reagents, calibrators and controls. Structurally unrelated compounds were evaluated by adding each substance to 6-Acetylmorphine spiked at low (7.5 ng/mL) and high (12.5 ng/mL) controls at the concentrations indicated. As shown in the tables below, all the pharmacologic compounds evaluated exhibited no cross-reactivity at the concentrations tested.

• Samples were tested in replications of five (n=5) in both qualitative and semiquantitative modes.

Interferents	Spiked Concentration (ng/mL)	Spiked 6-Acetylmorphine Level	
		Low Control	High Control
10,11 Dihydrocarbamazepine	85,000	Neg	Pos
11-nor-delta9-THC-COOH	10,000	Neg	Pos
Acetaminophen	500,000	Neg	Pos
Acetylsalicylic Acid	500,000	Neg	Pos
Amitriptyline	50,000	Neg	Pos
Amoxicillin	500,000	Neg	Pos
Amphetamine	100,000	Neg	Pos
Amisulpride	100,000	Neg	Pos
Benztropine Mesylate	50,000	Neg	Pos
Benzoylecgonine	100,000	Neg	Pos
Brompheniramine	75,000	Neg	Pos
Caffeine	500,000	Neg	Pos
Captopril	500,000	Neg	Pos
Chlordiazepoxide	100,000	Neg	Pos
Chlorpromazine	10,000	Neg	Pos
Clomipramine	150,000	Neg	Pos
Chloroquine	500,000	Neg	Pos
Cimetidine	500,000	Neg	Pos
Desipramine	50,000	Neg	Pos
Diazepam	100,000	Neg	Pos
Digoxin	100,000	Neg	Pos
Diphenhydramine	20,000	Neg	Pos
Doxepin HCl	10,000	Neg	Pos
Enalapril	500,000	Neg	Pos
Fluoxetine	500,000	Neg	Pos
Fluophenazine	500,000	Neg	Pos
Haloperidol	50,000	Neg	Pos

Hydroxychlroquine	100,000	Neg	Pos
Hydroxyzine	125,000	Neg	Pos
Ibuprofen	500,000	Neg	Pos
Imipramine	20,000	Neg	Pos
Levothyroxine	50,000	Neg	Pos
Methamphetamine	100,000	Neg	Pos
Maprotiline	500,000	Neg	Pos
Nalbuphine	100,000	Neg	Pos
Naproxen	500,000	Neg	Pos
Nortriptyline	175,000	Neg	Pos
Nifedipine	500,000	Neg	Pos
Nordiazepam	60,000	Neg	Pos
Oxazepam	100,000	Neg	Pos
Perphenazine	150,000	Neg	Pos
Phencyclidine	7,500	Neg	Pos
Phenobarbital	100,000	Neg	Pos
Procyclidine	150,000	Neg	Pos
Propoxyphene	25,000	Neg	Pos
Protriptyline	10,000	Neg	Pos
Ranitidine	500,000	Neg	Pos
Salicyluric Acid	500,000	Neg	Pos
Secobarbital	100,000	Neg	Pos
Sulpiride	500,000	Neg	Pos
Thioridazine	250,000	Neg	Pos
Triprolidine	125,000	Neg	Pos
Verapamil	500,000	Neg	Pos

Interference with Endogenous Substances

Compound	Spiked Concentration (mg/dL)	Spiked 6-Acetylmorphine Level	
		Low Control	High Control
Acetone	1000	Neg	Pos
Ascorbic acid	1500	Neg	Pos
Creatinine	500	Neg	Pos
Ethanol	1000	Neg	Pos
Galactose	10	Neg	Pos
Ɣ-globulin	500	Neg	Pos
Glucose	1000	Neg	Pos
Hemoglobin	300	Neg	Pos
Human serum albumin	500	Neg	Pos
Oxalic acid	100	Neg	Pos
Riboflavin	7.5	Neg	Pos
Sodium Chloride	6000	Neg	Pos
Urea	2000	Neg	Pos

Effect of pH: Drug free urine spiked with 7.5 or 12.5 ng/mL of 6-Acetylmorphine was adjusted to the indicated pH (3, 4, 5, 6, 7, 8, 9, 10, and 11). Samples were tested in replicates of five (n=5) in both qualitative and semiquantitative modes. The results demonstrated that the tested range of pH did not affect the performance of the candidate test.

Effect of Specific Gravity:
Urine that spanned a specific gravity range of 1.000–1.030 were spiked with 7.5 or 12.5 ng/mL of 6-Acetylmorphine (6-AM). Samples were tested in replicates of five (n=5) in both qualitative and semiquantitative modes. The results demonstrated that the specific gravity range evaluated did not affect the performance of the candidate test.

4. Assay Reportable Range:

   Not applicable.

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

   The device is traceable to a commercially available standard that was verified by LC/MS-MS or GC/MS.

6. Detection Limit:
   Not applicable.

7. Assay Cut-Off:
   See section VII A. 1

B Comparison Studies:

Method Comparison with Predicate Device: A Method comparison study was performed using one hundred and twenty-three unaltered clinical samples analyzed using the candidate test in one replicate, in both qualitative and semi-quantitative modes. The results were compared to LC-MS/MS (Liquid chromatography-tandem mass spectroscopy). The results obtained in the qualitative and semi-quantitative modes are summarized below.

Qualitative Mode:

CEDIA Heroin Metabolite (6-AM) Assay	< 50% of Cutoff concentration by LC-MS/MS (< 5ng/mL)	Near Cutoff Negative (Between 50% below the cutoff and the cutoff concentration as determined by LC- MS/MS) (5 – 9.9 ng/mL)	Near Cutoff Positive (Between the cutoff and 50% above the cutoff concentration as determined by LC-MS/MS) (10 – 15.0 ng/mL)	High Positives (Greater than 50% above cutoff concentration (> 15.0 ng/mL)
Positive	0	0	6	53
Negative	57	7	0	0

Semi-Quantitative Mode:

CEDIA Heroin Metabolite (6-AM) Assay	< 50% of Cutoff concentration by LC-MS/MS (< 5ng/mL)	Near Cutoff Negative (Between 50% below the cutoff and the cutoff concentration as determined by LC- MS/MS) (5 – 9.9 ng/mL)	Near Cutoff Positive (Between the cutoff and 50% above the cutoff concentration as determined by LC-MS/MS) (10 – 15.0 ng/mL)	High Positives (Greater than 50% above cutoff concentration (> 15.0 ng/mL)
Positive	0	1	5	53
Negative	57	6	1	0

1. Matrix Comparison:
   Not applicable.

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):

   None.

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.