

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE**

A. 510(k) Number:

k163101

B. Purpose for Submission:

New device

C. Measurand:

Buprenorphine

D. Type of Test:

Qualitative and semi-quantitative immunoassay

E. Applicant:

Microgenics Corporation

F. Proprietary and Established Names:

CEDIA Buprenorphine II Assay
CEDIA Buprenorphine II Calibrators
CEDIA Negative Calibrator II
CEDIA Buprenorphine II Control Set

G. Regulatory Information:

Regulation section	Classification	Product Code	Panel
21 CFR 862.3650	Class II	DJG	Toxicology (91)
21 CFR 862.3200	Class II	DLJ	Toxicology (91)
21 CFR 862.3280	Class I, reserved	LAS	Toxicology (91)

H. Intended Use:

1. Intended use(s):

Refer to Indications for Use below

2. Indication(s) for use:

The CEDIA® Buprenorphine II Assay is a homogeneous enzyme immunoassay for the qualitative and/or semiquantitative determination for the presence of buprenorphine and its metabolites in human urine at a cut-off concentration of 10 ng/mL. The assay is intended to be used in laboratories and provides a simple and rapid analytical screening procedure to detect buprenorphine in human urine. The assay is designed for use with a number of clinical chemistry analyzers.

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 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.

CEDIA® Buprenorphine II Calibrators:

The CEDIA® Buprenorphine II calibrators and CEDIA Negative Calibrator II are intended for the calibration of the CEDIA® Buprenorphine II Assay in human urine. For In Vitro Diagnostic Use Only.

CEDIA® Buprenorphine II Control Set:

The CEDIA® Buprenorphine II controls are used to validate the CEDIA® Buprenorphine II Assay calibration in human urine. For In Vitro Diagnostic Use Only.

3. Special conditions for use statement(s):

For prescription use only.

4. Special instrument requirements:

The CEDIA Buprenorphine II Assay is intended for use on automated clinical analyzers capable of maintaining a constant temperature, pipetting, mixing reagents, measuring enzymatic rates at 570 nm and timing the reaction accurately can be used to perform this immunoassay. All performance data was collected on a Beckman Coulter AU680 analyzer.

I. Device Description:

CEDIA® Buprenorphine II Assay is supplied as two liquid and two lyophilized reagent

kit homogeneous enzyme immunoassay:

- 1 EA Reconstitution Buffer
Contains buffer salts, mouse monoclonal anti-buprenorphine derivative antibody 0.8 - 1.0 mg/L, stabilizer, and preservative.
- 1a EA Reagent
Contains 0.171 g/L Enzyme Acceptor, buffer salts and preservative.
- 2 ED Reconstitution Buffer
Contains buffer salts, stabilizers, and preservatives
- 2a ED Reagent
Contains 0.175 mg/L Enzyme Donor conjugated to buprenorphine derivative, 1.67 g/L chlorophenol red- β -D-galactopyranoside, stabilizers, detergent and preservative.

The assay uses specific antibodies that can detect buprenorphine and its metabolites without significant cross-reactivity to other opiate compounds. In the assay, analyte in the sample competes with analyte conjugated to one inactive fragment of β -galactosidase for antibody binding site. If analyte is present in the sample, it binds to antibody, leaving the inactive enzyme fragments free to form active enzymes. If analyte is not present in the sample, antibody binds to analyte conjugated on the inactive fragment, inhibiting the reassociation of inactive β -galactosidase fragments, and no active enzyme is formed. The amount of active enzyme formed, and resultant absorbance change, is directly proportional to the amount of analyte present in the sample.

J. Substantial Equivalence Information:

1. Predicate device name(s):

Microgenics CEDIA Buprenorphine Assay

2. Predicate 510(k) number(s):

k040316

3. Comparison with predicate:

Similarities - Assay		
Item	Device	k040316 – Microgenics CEDIA Buprenorphine Assay
Intended Use	Same	Detection of buprenorphine in human urine
Methodology	Same	CEDIA (Cloned Enzyme

Similarities - Assay		
Item	Device	k040316 – Microgenics CEDIA Buprenorphine Assay
		Donor Immunoassay)
Intended Users	Same	Prescription users only
Reagents Form	Same	Lyophilized (requiring reconstitution) and liquid ready to use
Antibody	Same	Mouse monoclonal
Storage	Same	2 – 8° C until expiration date
Target Analyte	Same	Buprenorphine

Differences - Assay		
Item	Device	Predicate
Cutoff	10 ng/mL	5 ng/mL

Similarities – Calibrators		
Item	Device	Predicate
Form	Same	Liquid – ready to use
Storage	Same	2 – 8° C until expiration date

Differences - Calibrators		
Item	Device	Predicate
Calibrator Name	CEDIA® Buprenorphine II calibrators and controls	CEDIA® Buprenorphine calibrators and controls
Calibrator Levels	0, 10, 20, 50, 100 ng/mL	0, 5, 20, 50, 75 ng/mL

Similarities – Controls		
Item	Device	Predicate
Form	Same	Liquid – ready to use
Storage	Same	2 – 8° C until expiration date

Differences - Controls		
Item	Device	Predicate
Control Names	CEDIA® Buprenorphine II controls	CEDIA® Buprenorphine controls
Control Levels	7.5 and 12.5 ng/mL	3 and 7 ng/mL
Form	Same	Liquid – ready to use
Storage	Same	2 – 8° C until expiration date

K. Standard/Guidance Document Referenced (if applicable):

- CLSI EP05-A3 - Evaluation of Precision of Quantitative Measurement Procedures; Approved Guideline - Third Edition.
- CLSI EP06-A- Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline.
- CLSI EP07-A2 - Interference Testing In Clinical Chemistry; Approved Guideline - Second Edition.
- CLSI EP09-A3 - Measurement Procedure Comparison and Bias Estimation Using Patient Samples; Approved Guideline – Third Edition.
- CLSI EP25-A- Evaluation of Stability of In Vitro Diagnostic Reagents; Approved Guideline.

L. Test Principle:

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.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Precision was evaluated using CLSI Guideline EP05-A3 as a guideline, at one site with one analyzer, two operators, and two lots of reagents, calibrators and controls. 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 buprenorphine analyte to final concentrations of -100%, -75%,

-50%, -25%, below cutoff and +25%, +50%, +75% and +100%, above cutoff, and the concentrations were confirmed by LC-MS/MS. Results are summarized below:

Qualitative Mode – Lot 1

% of Cutoff	Target Conc. (ng/mL)	Measured Conc. (ng/mL)	# of determinants	# Negative/ # Positive
-100	0	0	80	80/0
-75	2.5	2.99	80	80/0
-50	5	5.31	80	80/0
-25	7.5	7.63	80	80/0
100	10	10.99	80	18/62
+25	12.5	12.97	80	0/80
+50	15	15.05	80	0/80
+75	17.5	18.92	80	0/80
+100	20	20.38	80	0/80

Qualitative Mode – Lot 2

% of Cutoff	Target Conc. (ng/mL)	Measured Conc. (ng/mL)	# of determinants	# Negative/ # Positive
-100	0	0	80	80/0
-75	2.5	2.99	80	80/0
-50	5	5.31	80	80/0
-25	7.5	7.63	80	80/0
100	10	10.99	80	27/53
+25	12.5	12.97	80	0/80
+50	15	15.05	80	0/80
+75	17.5	18.92	80	0/80
+100	20	20.38	80	0/80

Semi-Quantitative Mode – Lot 1

% of Cutoff	Target Conc. (ng/mL)	Measured Conc. (ng/mL)	# of determinants	# Negative/ # Positive
-100	0	0	80	80/0
-75	2.5	2.99	80	80/0
-50	5	5.31	80	80/0
-25	7.5	7.63	80	80/0
100	10	10.99	80	7/73
+25	12.5	12.97	80	0/80
+50	15	15.05	80	0/80
+75	17.5	18.92	80	0/80
+100	20	20.38	80	0/80

Semi-Quantitative Mode – Lot 2

% of Cutoff	Target Conc. (ng/mL)	Measured Conc. (ng/mL)	# of determinants	# Negative/ # Positive
-100	0	0	80	80/0
-75	2.5	2.99	80	80/0
-50	5	5.31	80	80/0
-25	7.5	7.63	80	80/0
100	10	10.99	80	35/45
+25	12.5	12.97	80	0/80
+50	15	15.05	80	0/80
+75	17.5	18.92	80	0/80
+100	20	20.38	80	0/80

b. Linearity/assay reportable range:

The sponsor performed a spike recovery study using two lots each of reagent, calibrators and controls and analyzed concentrations of 7.5, 10, and 12.5 ng/mL. Samples were analyzed in semi-quantitative mode in replicates of 5. Recoveries ranged from 96.6% - 105.3%.

The sponsor also performed a linearity study using two lots each of reagent, calibrators and controls and analyzed concentrations of 0, 5, 10, 20, 30, 40, 50, 60, 70, 80, 90, and 100 ng/mL. Samples were analyzed in semi-quantitative mode in replicates of 5. Recoveries ranged from a low of 98.3% to a high of 119.8%. Recovery at the claimed cutoff of 10 ng/mL was 109.7%.

c. Traceability, Stability, Expected values (controls, calibrators, or methods):

Traceability: The primary calibrators and controls are traceable to a commercially available Buprenorphine drug stock with a starting concentration of 1 mg/mL.

Value Assignment: The nominal values for calibrators are 0, 10, 20, 50 and 100 ng/mL, and 7.5 and 12.5 ng/mL for controls. All values are verified by LC-MS/MS.

The sponsor's protocols and acceptance criteria were reviewed and found to be acceptable.

Stability: Real time and accelerated stability studies for both controls and calibrators were conducted. Protocols and acceptance criteria were reviewed and found to be acceptable. The results support the manufacturer's stability claims of 60 days for an opened vial and 18 months for an unopened vial for both calibrators and controls. Real time studies are ongoing.

d. *Detection limit:*

Not applicable.

e. *Analytical specificity:*

Buprenorphine and metabolites

To evaluate cross-reactivity, drug-free urine was spiked with norbuprenorphine, buprenorphine- β -D-glucuronide and norbuprenorphine- β -D glucuronide. Percent cross-reactivity was calculated as (Cut-off concentration / Lowest concentration of cross reactant that gives a positive result) x 100. Results are summarized below:

Compound	Lowest concentration producing a positive result (ng/mL)	Percent cross-reactivity
Buprenorphine	10	100
Norbuprenorphine	8	125
Buprenorphine- β -D-glucuronide	13	77
Norbuprenorphine- β -D-glucuronide	10	100

Opiates and Structurally Related Compounds

The following opiates and structurally related compounds were analyzed and found to have a cross-reactivity of <0.01%.

Compound	Highest Concentration Tested	Result
6-Acetyl morphine	100,000	Negative
Diacetylmorphine (Heroin)	100,000	Negative
Codeine	100,000	Negative
Dextromethorphan	100,000	Negative
Dihydrocodeine	100,000	Negative
EDDP (2-ethylidene-1,5-dimethyl-3,3-diphenylpyrrolidine)	100,000	Negative
EMDP ((2-Ethyl-5-methyl-3,3-	100,000	Negative

Compound	Highest Concentration Tested	Result
diphenylpyrroline)		
Fentanyl	100,000	Negative
Hydrocodone	100,000	Negative
Hydromorphone	100,000	Negative

The following opiates and structurally related compounds were analyzed and found to have a cross-reactivity of <0.1%.

Compound	Highest Concentration Tested	Result
Hydromorphone-β-Dglucuronide	10,000	Negative
Oxymorphone-β-Dglucuronide	10,000	Negative

Structurally unrelated compounds

Interference from structurally unrelated compounds was evaluated by spiking these compounds into urine samples containing near cutoff negative (7.5 ng/mL) and near cutoff positive (12.5 ng/mL) concentrations of buprenorphine. The compounds listed in the table below did not cause any positive or negative interference at the concentrations shown:

Compound	Concentration tested
Acetaminophen	500,000
Acetylsalicylic acid	500,000
Amitriptyline	50,000
Amoxicillin	100,000
Amphetamine	1,000,000
Amisulpride	100,000
Benzoyllecgonine	1,000,000
Caffeine	100,000
Carbamazepine	100,000
Chlorpromazine	100,000
Clomipramine	25,000
Chloroquine	100,000
Cimetidine	500,000
Desipramine	10,000
Doxepine	25,000
Diphenylhydramine	100,000
Ephedrine	100,000

Compound	Concentration tested
Fluoxetine	100,000
Fluphenazine	100,000
Hydroxychloroquine	100,000
Ibuprofen	100,000
Imipramine	25,000
Maprotiline	100,000
Mitragynine	100,000
7-OH Mitragynine	10,000
Nalbuphine	100,000
Nortryptiline	50,000
Oxazepam	100,000
Phencyclidine	100,000
Phenobarbital	100,000
Ranitidine	500,000
Secobarbital	100,000
Sulpiride	100,000
Thioridazine	100,000
Trimipramine	25,000

Endogenous compounds

Potential interference from endogenous compounds was evaluated by spiking these compounds into urine samples containing near cutoff negative (7.5 ng/mL) and near cutoff positive (12.5 ng/mL) concentrations of buprenorphine. The compounds or conditions listed in the table below did not cause any positive or negative interference at the concentrations shown:

Compounds	Tested Conc. (mg/dL)
Negative Urine	0
Acetaminophen	10
Acetone	500
Acetylsalicylic Acid	10
Ascorbic Acid	150
Caffeine	10
Creatinine	400
Ethanol	10
Galactose	5
Glucose	1000
Hemoglobin	150
Human Serum Albumin	200
Ibuprophen	10
Oxalic acid	50

Compounds	Tested Conc. (mg/dL)
Riboflavin	3
Sodium Chloride	1000
Urea	1000

Specific gravity and pH

Interference from specific gravity and pH was evaluated by adjusting the specific gravity and pH of samples with near cutoff negative (7.5 ng/mL) and near cutoff positive (12.5 ng/mL) concentrations of buprenorphine. The following specific gravity or pH levels did not cause any positive or negative interference:

Specific gravity of 1.002, 1.004, 1.008, 1.013, 1.016, 1.018, 1.022, 1.023, 1.025, and 1.030.

pH of 3, 4, 5, 6, 7, 8, 9, 10, and 11

f. Assay cut-off:

Analytical performance of the device around the claimed cutoff is described in precision section (1 a.) above.

2. Comparison studies:

a. Method comparison with predicate device:

Candidate Device Results vs. stratified LC-MS/MS Values – Semi-quantitative

Candidate Device Results	Negative or less than half the cutoff concentration by LC-MS/MS analysis	Near Cutoff Negative (Between 50% below the cutoff and the cutoff concentration by LC-MS/MS analysis)	Near Cutoff Positive (Between the cutoff and 50% above the cutoff concentration by LC-MS/MS analysis)	High Positive (greater than 50% above the cutoff concentration by LC-MS/MS analysis)
Positive	43	4	5	45
Negative	50	6	0	0

LC-MS/MS values used to categorize samples in this table are based on the concentration of buprenorphine found in the sample.

% Agreement among positives is 50/50 = 100%

% Agreement among negatives is 56/103 = 54%

Candidate Device Results vs. stratified LC-MS/MS Values – Qualitative

Candidate Device Results	Negative or less than half the cutoff concentration by LC-MS/MS analysis	Near Cutoff Negative (Between 50% below the cutoff and the cutoff concentration by LC-MS/MS analysis)	Near Cutoff Positive (Between the cutoff and 50% above the cutoff concentration by LC-MS/MS analysis)	High Positive (greater than 50% above the cutoff concentration by LC-MS/MS analysis)
Positive	42	4	5	45
Negative	51	6	0	0

LC-MS/MS values used to categorize samples in this table are based on the concentration of buprenorphine found in the sample.

% Agreement among positives is 50/50 = 100%

% Agreement among negatives is 57/103 = 55%

Summary of discordant results

Sample ID	Qual	Semi-Quant (ng/mL)	Bup	NorBup	BupGlu	NorBup Gluc
51	Pos	10.08	<0.65*	2.27	1.96	6.18
52	Pos	10.02	<0.65*	0.69	3.15	6.84
53	Neg	10.42	<0.65*	1.08	7.89	1.82
54	Pos	11.59	<0.65*	1.09	5.67	5.54
55	Pos	10.40	<0.65*	3.27	2.54	7.92
56	Pos	16.36	<0.65*	4.02	7.46	3.73
57	Pos	17.31	<0.65*	3.28	10.67	3.09
58	Pos	19.82	<0.65*	5.03	10.91	2.05
59	Pos	18.73	<0.65*	3.10	9.09	6.59
60	Pos	22.63	<0.65*	4.18	8.30	7.34
61	Pos	18.95	<0.65*	1.96	9.90	9.90
62	Pos	26.11	<0.65*	4.36	10.87	6.92
63	Pos	24.99	<0.65*	5.26	8.41	9.01
64	Pos	24.91	<0.65*	3.86	23.19	<0.65*
65	Pos	20.87	<0.65*	1.44	14.06	14.06
66	Pos	23.21	<0.65*	2.23	25.24	2.50
67	Pos	30.27	<0.65*	4.42	8.82	16.84
68	Pos	31.35	<0.65*	16.52	9.41	5.47

Sample ID	Qual	Semi-Quant (ng/mL)	Bup	NorBup	BupGlu	NorBup Gluc
69	Pos	35.38	<0.65*	7.13	5.30	22.38
70	Pos	40.38	<0.65*	12.21	18.65	9.11
71	Pos	38.44	<0.65*	2.93	12.40	28.84
72	Pos	48.60	<0.65*	23.41	15.34	5.44
73	Pos	62.31	<0.65*	5.47	36.52	25.00
74	Pos	81.31	<0.65*	33.59	23.42	12.72
75	Pos	88.67	<0.65*	26.22	32.43	23.1
76	Pos	79.26	<0.65*	6.34	80.00	2.77
77	Pos	>100	<0.65*	8.63	56.89	46.95
78	Pos	>100	<0.65*	101.98	10.40	9.90
79	Pos	>100	<0.65*	7.91	26.43	144.00
80	Pos	>100	<0.65*	49.66	97.61	121.12
81	Pos	>100	<0.65*	<0.65*	145.72	394.81
82	Pos	>100	<0.65*	129.95	105.07	664.47
83	Pos	>100	0.81	32.14	39.52	59.14
84	Pos	63.54	0.86	7.41	29.46	31.38
85	Pos	20.48	0.90	5.42	11.54	<0.65*
86	Pos	>100	0.91	54.00	18.10	10.52
87	Pos	46.32	2.00	12.03	13.58	16.24
88	Pos	>100	2.00	6.83	193.42	131.65
89	Pos	>100	2.02	75.75	174.74	442.98
90	Pos	66.32	2.48	6.53	57.67	1.52
91	Pos	>100	3.63	80.26	733.7	624.02
92	Pos	>100	4.38	69.28	146.16	349.33
93	Pos	>100	4.45	59.03	55.01	17.31
100	Pos	>100	8.64	36.91	>1000**	224.42
101	Pos	>100	8.94	51.32	497.32	55.06
102	Pos	>100	5.22	35.13	85.99	22.24
103	Pos	77.36	6.60	147.58	195.67	40.28

Abbreviations: Bup – Buprenorphine, NorBup – Norbuprenorphine, BupGlu – Buprenorphine Glucuronide, NorBupGluc – Norbuprenorphine Glucuronide

*0.65 ng/mL is the lower limit of quantitation for the buprenorphine, norbuprenorphine, buprenorphine-glucuronide and norbuprenorphine-glucuronide LC-MS/MS assays

****1000 ng/mL is the upper limit of quantitation for the buprenorphine, norbuprenorphine, buprenorphine-glucuronide and norbuprenorphine-glucuronide LC-MS/MS assays.**

b. Matrix comparison:

Not applicable. This assay is intended to be used with urine samples only.

3. Clinical studies:

a. Clinical Sensitivity:

Not applicable.

b. Clinical specificity:

Not applicable.

c. Other clinical supportive data (when a. and b. are not applicable):

Not applicable.

4. Clinical cut-off:

Not applicable.

5. Expected values/Reference range:

Not applicable.

N. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

O. Conclusion:

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